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 NEWS  1          Web Page URLs for STN Seminar Schedule - N. America
 NEWS  2          "Ask CAS" for self-help around the clock
 NEWS  3  JUL 12  BEILSTEIN enhanced with new display and select options,
                  resulting in a closer connection to BABS
 NEWS  4  AUG 02  IFIPAT/IFIUDB/IFICDB reloaded with new search and display
                  fields
 NEWS  5  AUG 02  CAplus and CA patent records enhanced with European and Japan
                  Patent Office Classifications
 NEWS  6  AUG 02  The Analysis Edition of STN Express with Discover!
                  (Version 7.01 for Windows) now available
 NEWS  7  AUG 27  BIOCOMMERCE: Changes and enhancements to content coverage
 NEWS  8  AUG 27  BIOTECHABS/BIOTECHDS: Two new display fields added for legal
                  status data from INPADOC
 NEWS  9  SEP 01  INPADOC: New family current-awareness alert (SDI) available
 NEWS 10  SEP 01  New pricing for the Save Answers for SciFinder Wizard within
                  STN Express with Discover!
 NEWS 11  SEP 01  New display format, HITSTR, available in WPIDS/WPINDEX/WPIX
 NEWS 12  SEP 27  STANDARDS will no longer be available on STN
 NEWS 13  SEP 27  SWETSCAN will no longer be available on STN
 NEWS 14  OCT 28  KOREAPAT now available on STN

 NEWS EXPRESS  OCTOBER 29 CURRENT WINDOWS VERSION IS V7.01A, CURRENT
               MACINTOSH VERSION IS V6.0c(ENG) AND V6.0Jc(JP),
               AND CURRENT DISCOVER FILE IS DATED 11 AUGUST 2004
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FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004

=> file medline biosis caplus embase wpids
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                       ENTRY    SESSION
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FULL ESTIMATED COST 0.42 0.42

FILE 'MEDLINE' ENTERED AT 15:53:29 ON 18 NOV 2004

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FILE 'WPIDS' ENTERED AT 15:53:29 ON 18 NOV 2004
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=> Shiga 5A toxin
L1 0 SHIGA 5A TOXIN

=> (Shiga (5A) toxin) (s) (resistant or resistance or insensitive or (not (w)sensitive))
MISSING TERM 'OR (NOT'
The search profile entered contains a left parenthesis,
'(' followed by an operator.

=> (Shiga (5A) toxin) (s) (resistant or resistance or insensitive or ("not" (w)sensitive))
L2 193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE OR ("NOT" (W) SENSITIVE))

=> d scan

L2 193 ANSWERS CAPLUS COPYRIGHT 2004 ACS on STN
CC 10-0 (Microbial, Algal, and Fungal Biochemistry)
Section cross-reference(s): 17
TI Antimicrobial resistance of foodborne pathogens
ST review antimicrobial resistance foodborne pathogen
IT Antibiotic resistance
Antibiotics
Campylobacter
Food contamination
Human
Listeria monocytogenes
Pathogenic bacteria
Public health
Salmonella
Yersinia
(antibiotic resistance of foodborne pathogens and transmission to humans via food)
IT Escherichia coli
(**shiga toxin** producing; antibiotic **resistance** of foodborne pathogens and transmission to humans via food)

HOW MANY MORE ANSWERS DO YOU WISH TO SCAN? (1):3

L2 193 ANSWERS BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI EVALUATION OF THE ROLE OF SHIGA AND SHIGA-LIKE TOXINS IN MEDIATING DIRECT DAMAGE TO HUMAN VASCULAR ENDOTHELIAL CELLS.
IT Miscellaneous Descriptors

SHIGELLA-DYSENTERIAE ESCHERICHIA-COLI MEMBRANE

L2 193 ANSWERS BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Characterization of a **shiga-toxin** 1-**resistant** stock of Vero cells.
IT Methods & Equipment
MTT assay [methylthiazolyldiphenyl-tetrazolium bromide assay]: laboratory techniques; binding assay: laboratory techniques; confocal microscopy: imaging and microscopy techniques, laboratory techniques; lipid analysis: laboratory techniques
IT Miscellaneous Descriptors
cell viability; intracellular transport; serum depression

L2 193 ANSWERS CAPLUS COPYRIGHT 2004 ACS on STN
CC 10-6 (Microbial, Algal, and Fungal Biochemistry)
Section cross-reference(s): 14
TI Detection and antibiotic **resistance** of **Shiga** **toxin** gene positive E. coli isolated from rectal swabs of children with diarrhea in Slovakia
ST **Shiga toxin** Escherichia detection antibiotic **resistance**; gene Shiga toxin Escherichia
IT Escherichia coli
(STEC; detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)
IT Toxins
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(Shiga; detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)
IT Antibiotic **resistance**
Diagnosis
Feces
Human
PCR (polymerase chain reaction)
Virulence (microbial)
(detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)
IT Gene, microbial
RL: ANT (Analyte); BSU (Biological study, unclassified); ANST (Analytical study); BIOL (Biological study)
(stx1; detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)
IT Gene, microbial
RL: ANT (Analyte); BSU (Biological study, unclassified); ANST (Analytical study); BIOL (Biological study)
(stx2; detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)
IT 69-53-4, Ampicillin 8025-81-8, Spiramycin. 79198-29-1, Amoxicillin/clavulanic acid
RL: PAC (Pharmacological activity); BIOL (Biological study)
(detection and antibiotic **resistance** of **Shiga** **toxin** gene-pos. Escherichia coli isolated from rectal swabs of children with diarrhea in Slovakia)

HOW MANY MORE ANSWERS DO YOU WISH TO SCAN? (1):end

=> d his

(FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18 NOV 2004

L1 0 SHIGA 5A TOXIN
L2 193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE

=> ((mutated or mutant) (s) subunit) and l2
L3 1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2

=> d ibib abs l3

L3 ANSWER 1 OF 1 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
ACCESSION NUMBER: 2000-412091 [35] WPIDS
DOC. NO. CPI: C2000-124883
TITLE: Expression cassette used as live vector vaccine comprises nucleotide sequence encoding origin of replication and plasmid maintenance system which includes a post-segregational killing and a partitioning function.
DERWENT CLASS: B04 C06 D16
INVENTOR(S): GALEN, J E
PATENT ASSIGNEE(S): (UYMA-N) UNIV MARYLAND BALTIMORE
COUNTRY COUNT: 87
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000032047	A1	20000608	(200035)*	EN	127
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ TZ UG ZW					
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US VN YU ZA ZW					
AU 2000020364	A	20000619	(200044)		
EP 1135025	A1	20010926	(200157)	EN	
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI					
NO 2001002721	A	20010731	(200157)		
CZ 2001001538	A3	20011114	(200175)		
HU 2001004609	A2	20020328	(200234)		
ZA 2001005383	A	20020424	(200237)		135
US 6413768	B1	20020702	(200248)		
MX 2001005449	A1	20011201	(200282)		
JP 2003506007	W	20030218	(200315)		170
US 6703233	B1	20040309	(200418)		
NZ 511449	A	20040528	(200437)		
US 2004161420	A1	20040819	(200455)		

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000032047	A1	WO 1999-US28499	19991202
AU 2000020364	A	AU 2000-20364	19991202
EP 1135025	A1	EP 1999-964042	19991202
		WO 1999-US28499	19991202
NO 2001002721	A	WO 1999-US28499	19991202
		NO 2001-2721	20010601
CZ 2001001538	A3	WO 1999-US28499	19991202
		CZ 2001-1538	19991202
HU 2001004609	A2	WO 1999-US28499	19991202

ZA 2001005383	A		HU 2001-4609	19991202
US 6413768	B1		ZA 2001-5383	20010629
MX 2001005449	A1		US 1998-204117	19981202
JP 2003506007	W		MX 2001-5449	20010531
			WO 1999-US28499	19991202
US 6703233	B1	CIP of Provisional	JP 2000-584755	19991202
			US 1998-204117	19981202
			US 1999-158738P	19991012
NZ 511449	A		US 1999-453313	19991202
			NZ 1999-511449	19991202
US 2004161420	A1	CIP of Provisional Div ex	WO 1999-US28499	19991202
			US 1998-204117	19981202
			US 1999-158738P	19991012
			US 1999-453313	19991202
			US 2004-750965	20040105

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2000020364	A	Based on	WO 2000032047
EP 1135025	A1	Based on	WO 2000032047
CZ 2001001538	A3	Based on	WO 2000032047
HU 2001004609	A2	Based on	WO 2000032047
JP 2003506007	W	Based on	WO 2000032047
US 6703233	B1	CIP of	US 6413768
NZ 511449	A	Div in	NZ 529508
		Based on	WO 2000032047
US 2004161420	A1	CIP of	US 6413768
		Div ex	US 6703233

PRIORITY APPLN. INFO: US 1999-158738P 19991012; US 1998-204117 19981202; US 1999-453313 19991202; US 2004-750965 20040105

AN 2000-412091 [35] WPIDS

AB WO 200032047 A UPAB: 20000725

NOVELTY - An independently functioning expression cassette (I), comprises a nucleotide sequence encoding an origin of replication (ORI) and a nucleotide sequence encoding a plasmid maintenance system (PMS) which includes a post-segregational killing function (PSK) and a partitioning function.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) an amplifiable plasmid replicon (II) comprising (I);

(2) a bacterial cell comprising (II);

(3) an attenuated bacterial live vector vaccine (III), comprising a bacterial species containing a replicon comprising a nucleotide sequence encoding an antigen of interest, and a nucleotide sequence encoding a PMS;

(4) a conditionally unstable plasmid (IV), for examining changes in plasmid stability resulting from incorporation of plasmid maintenance system, comprises an ORI yielding an average copy number of 2-75 copies and a promoter driving the expression of a protein or peptide and whose over expression imposes a metabolic burden on a bacterium, which favors plasmid loss;

(5) making (M1) a stabilized (III), which involves transforming a bacterial live vector with a replicon comprising a PMS which includes one PSK and one partitioning function, and a nucleotide sequence encoding one or more antigen;

(6) a DNA (V), comprising a modified ompC promoter phenotypically characterized so that the promoter exhibits higher rates of osmotically regulated expression in relation to a corresponding non-mutated ompC

promoter; and

(7) an expression plasmid (VI) comprising (V).

ACTIVITY - Cytostatic; antibacterial; virucide; hepatropic; antiinflammatory; immunosuppressive; dermatological; antiasthmatic; antiallergic; neuroprotective; antiarthritic; antirheumatic; No supporting data is given.

MECHANISM OF ACTION - Vaccine.

USE - (IV) is used for eliciting an immune response in a human or bovine subject (claimed). (I) is used for transforming a bacterial cell which is cultured, and transformed into a subject to elicit an immune response. (I) can also be used to vaccinate a subject against Salmonella typhi. (I) may comprise an antigen for hepatitis B, Haemophilus influenzae type b, hepatitis A, acellular pertussis (acP), varicella, rotavirus, Streptococcus pneumoniae, or Neisseria meningitidis, and can be used as vaccines against diseases caused by these agents. (I) can be also used as a cancer vaccine. The antigens encoded by the plasmids are designed to provoke an immune response to autoantigens, B cell receptors and/or T cell receptors which are implicated in autoimmune or immunological diseases. Where an inappropriate immune response is raised against body tissues, or environmental antigens, the vaccines may immunize against the autoantigens, B cell receptors and/or T cell receptors to modulate the responses and ameliorate diseases, such as myasthenia gravis, lupus erythematosis, rheumatoid arthritis, multiple sclerosis, allergies and asthma.

ADVANTAGE - The plasmid maintenance systems incorporated into multicopy expression plasmids encoding one or more proteins or peptides of interest, enhances the level of expression of the protein or peptide of interest. The plasmid maintenance systems provide improved stability of recombinant plasmids, overcoming prior art problems of plasmid instability.

DESCRIPTION OF DRAWING(S) - The figure shows the pGEN expression plasmid pGEN2.

Dwg.1/8

```
=> ((mutated or mutant) (s) subunit)
L4       20901 ((MUTATED OR MUTANT) (S) SUBUNIT)

=> d his

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     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1             0 SHIGA 5A TOXIN
L2           193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3             1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4         20901 ((MUTATED OR MUTANT) (S) SUBUNIT)

=> (mutated or mutant or virulent) (s) (Shiga (5A) toxin)
L5           182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)

=> (mutated or mutant or virulent) (s) (Shiga (w) toxin)
L6           122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)

=> L2 and L6
L7            16 L2 AND L6

=> dup rem l7
PROCESSING COMPLETED FOR L7
L8            10 DUP REM L7 (6 DUPLICATES REMOVED)
```

=> t ti l8 1-10

L8 ANSWER 1 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Construction, characterization, and animal testing of WRSd1, a Shigella dysenteriae 1 vaccine.

L8 ANSWER 2 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Involvement of surface polysaccharides in the organic acid **resistance** of **Shiga Toxin**-producing Escherichia coli O157:H7.

L8 ANSWER 3 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Ferrets as a model system for renal disease secondary to intestinal infection with Escherichia coli O157:H7 and other Shiga toxin-producing E. coli.

L8 ANSWER 4 OF 10 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Sanitation of produce including fruits and/or vegetables involves applying bacteriophage(s) to the produce.

L8 ANSWER 5 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Flavopiridol induces apoptosis and caspase-3 activation of a newly characterized Burkitt's lymphoma cell line containing mutant p53 genes.

L8 ANSWER 6 OF 10 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Expression cassette used as live vector vaccine comprises nucleotide sequence encoding origin of replication and plasmid maintenance system which includes a post-segregational killing and a partitioning function.

L8 ANSWER 7 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Shiga toxin-producing Escherichia coli isolates from cases of human disease show enhanced adherence to intestinal epithelial (Henle 407) cells.

L8 ANSWER 8 OF 10 MEDLINE on STN DUPLICATE 1
TI Isolation of **Shiga toxin-resistant** Vero cells and their use for easy identification of the toxin.

L8 ANSWER 9 OF 10 MEDLINE on STN DUPLICATE 2
TI Deletion of the **Shiga toxin** gene in a chlorate-**resistant** derivative of Shigella dysenteriae type 1 that retains virulence.

L8 ANSWER 10 OF 10 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Localization of stx, a determinant essential for high-level production of shiga toxin by Shigella dysenteriae serotype 1, near pyrF and generation of stx transposon mutants.

=> d ibib abs l8, 8

L8 ANSWER 8 OF 10 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 88314296 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3045003
TITLE: Isolation of **Shiga toxin-resistant** Vero cells and their use for easy

identification of the toxin.

AUTHOR: Kongmuang U; Honda T; Miwatani T
CORPORATE SOURCE: Research Institute for Microbial Diseases, Osaka University, Japan.
SOURCE: Infection and immunity, (1988 Sep) 56 (9) 2491-4. Journal code: 0246127. ISSN: 0019-9567.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198810
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19900308
Entered Medline: 19881004

AB **Shiga toxin-resistant** Vero cells were isolated by treatment of the cells with nitrosoguanidine. These **mutant** cells were not affected by **Shiga toxin** at more than 1 microgram/ml, although the parent Vero cells were sensitive to 25 pg of the toxin per ml. Immunofluorescence studies showed that all the mutant cells had lost toxin-binding capacity. The cytotoxic activities of various bacterial cultures against the parent and mutant cells were compared. All samples from 10 strains of Shigella dysenteriae type 1 and all three strains of Escherichia coli O157:H7 tested showed cytotoxicity to the parent cells but not to the mutant cells. Samples from other organisms, such as Shigella flexneri, Shigella sonnei, Clostridium difficile, Aeromonas hydrophila, Aeromonas sobria, and other E. coli strains, either had no effect or were cytotoxic on both the parent and mutant cells. Thus, these mutant cells could be used to identify Shiga-like toxin and distinguish it from other cytotoxins. The results also suggest the presence of a receptor for Shiga-like toxin on Vero cells that is essential for expression of the cytotoxicity of Shiga toxin but is not essential for growth of Vero cells.

=> FIL STNGUIDE
COST IN U.S. DOLLARS

	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	82.61	83.03

FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

FILE CONTAINS CURRENT INFORMATION.
LAST RELOADED: Nov 12, 2004 (20041112/UP).

=> file medline biosis caplus embase wpids
COST IN U.S. DOLLARS

	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	0.06	83.09

FILE 'MEDLINE' ENTERED AT 16:08:34 ON 18 NOV 2004

FILE 'BIOSIS' ENTERED AT 16:08:34 ON 18 NOV 2004

FILE 'CAPLUS' ENTERED AT 16:08:34 ON 18 NOV 2004

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FILE 'WPIDS' ENTERED AT 16:08:34 ON 18 NOV 2004
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=> d his

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     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1            0 SHIGA 5A TOXIN
L2          193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3            1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4        20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5          182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6          122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7           16 L2 AND L6
L8           10 DUP REM L7 (6 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18
     NOV 2004

=> (mutated or mutant) (w) ((virulent or virulence or toxicity or toxic or
cytotoxic or cytotoxicity) 3N (increas? or more or great?))
MISSING OPERATOR TOXICITY) 3N
The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> (mutated or mutant) (w) ((virulent or virulence or toxicity or toxic or
cytotoxic or cytotoxicity) (3N) (increas? or more or great?))
L9            2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
                TOXIC OR CYTOTOXIC OR CYTOTOXICITY) (3N) (INCREAS? OR MORE OR
                GREAT?))

=> dup rem L9
PROCESSING COMPLETED FOR L9
L10           2 DUP REM L9 (0 DUPLICATES REMOVED)

=> t ti L10 1-2

L10  ANSWER 1 OF 2  WPIDS  COPYRIGHT 2004 THE THOMSON CORP on STN
TI   New isolated mutated human p53 polypeptides for inducing toxicity in a
     cell, treating cancer and identifying compounds that mimic toxic or
     supertransactivating mutations.

L10  ANSWER 2 OF 2  CAPLUS  COPYRIGHT 2004 ACS on STN
TI   Characterization of lectins in the cotton-Verticillium dahliae Kleb.
     system


=> d ibib abs l10 1-2

L10  ANSWER 1 OF 2  WPIDS  COPYRIGHT 2004 THE THOMSON CORP on STN
ACCESSION NUMBER:     2001-123321 [13]   WPIDS
DOC. NO. CPI:         C2001-035890
TITLE:                New isolated mutated human p53 polypeptides for inducing
                      toxicity in a cell, treating cancer and identifying
                      compounds that mimic toxic or supertransactivating
```

mutations.
DERWENT CLASS: B04 D16
INVENTOR(S): INGA, A; RESNICK, M A
PATENT ASSIGNEE(S): (USSH) US DEPT HEALTH & HUMAN SERVICES
COUNTRY COUNT: 95
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2001009325	A2	20010208	(200113)*	EN	144
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW					
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW					
AU 2000062395	A	20010219	(200129)		
EP 1204745	A2	20020515	(200239)	EN	
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI					
JP 2003506041	W	20030218	(200315)		151

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2001009325	A2	WO 2000-US20538	20000728
AU 2000062395	A	AU 2000-62395	20000728
EP 1204745	A2	EP 2000-948979	20000728
		WO 2000-US20538	20000728
JP 2003506041	W	WO 2000-US20538	20000728
		JP 2001-514117	20000728

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2000062395	A	Based on	WO 2001009325
EP 1204745	A2	Based on	WO 2001009325
JP 2003506041	W	Based on	WO 2001009325

PRIORITY APPLN. INFO: US 1999-146634P 19990730
AN 2001-123321 [13] WPIDS
AB WO 200109325 A UPAB: 20010307
NOVELTY - Isolated polypeptides (I) of human p53 containing specific mutations, are new.

DETAILED DESCRIPTION - New isolated polypeptides (I) of p53 having the following residues containing the mutations (in brackets):

(a) 117 to 172 (V122A);
(b) 272 to 282 (C277W);
(c) 272 to 282 (C277R);
(d) 333 to 343 (F338L);
(e) 153 to 163 (V157I;
(f) 70 to 80 (A76T);
(g) 145 to 155 (T150A);
(h) 115 to 125 (S121C);
(i) 90 to 100 (S96P);
(j) 110 to 120 (H115R);
(k) 120 to 130 (C124Y);
(l) 115 to 125 (S121F);
(m) 118 to 128 (T123A);
(n) 120 to 130 (C124F);

(o) 235 to 245 (S240N);
(p) 110 to 120 (S116T);
(q) 340 to 350 (N345S);
(r) 118 to 128 (T123S);
(s) 180 to 190 (D184G);
(t) 283 to 293 (N288K);
(u) 193 to 203 (E198V);
(v) 110 to 120 (H115R);
(w) 85 to 95 (W92R);
(x) 90 to 100 (S96P);
(y) 110 to 120 (S116T);
(z) 225 to 235 (N228K);
(a') 113 to 123 (T118A);
(b') 118 to128 (T123P);
(c') 132 to 142 (L137R);
(d') 155 to 165 (M160T);
(e') 235 to 245 (N239Y);
(f') 280 to 290 (E285A);
(g') 50 to 150 (A76T) and (V22A);
(h') 50 to 200 (W91C), (C124R), (Q136K), and (T150A); or
(i') 100 to 200 (C124R), (Q136K) and (T150A).

INDEPENDENT CLAIMS are also included for the following:

(1) an isolated nucleic acid (II) encoding (I);

(2) detecting (M1) a supertransactivating mutation in (II) comprising:

(a) introducing (II) comprising an inducible promoter containing GAL 1 linked to a human p53 coding sequence, into a yeast cell that has a reporter gene linked to a DNA sequence that p53 binds;

(b) plating the cell on raffinose as a carbon source; and

(c) identifying colonies on plates where wild type and mutant colonies yield different colored colonies;

(3) detecting (M2) a toxic mutation in the human p53 gene comprises (M1), where the cell can be plated on glucose, raffinose or galactose;

(4) detecting (M3) a toxic mutation in the human p53 gene comprising:

(a) introducing a nucleic acid encoding an unidentified human p53 and containing an on-off promoter linked to the coding sequence, into a yeast cell;

(b) incubating the cell in synthetic yeast medium in the presence and absence of an inducer for the promoter; and

(c) yeast expressing wildtype p53 yield grow in the presence or absence of the inducer and yeast expressing a mutation yield grow in the presence of the inducer;

(5) inducing (M4) toxicity in a cell by administering (I);

(6) screening (M5) for compounds that can mimic a toxic p53 mutation comprising:

(a) introducing into a yeast cell of (M1(a)), a nucleic acid that encodes a non-toxic or wildtype p53 and that contains an inducible promoter linked to the coding sequence;

(b) introducing the compound to the cell;

(c) plating the cell on glucose, raffinose or galactose;

(d) identifying a compound that mimics a toxic mutation preventing growth of colonies expressing wildtype or non-toxic mutant p53;

(7) screening (M6) for compounds that can mimic a toxic p53 mutation comprising:

(a) introducing into a yeast cell a nucleic acid which encodes a non-toxic mutant or wildtype p53 and comprising an on-off promoter linked to the coding sequence;

(b) introducing the compound to the cell;

(c) (M3(b)); and

(d) identifying a compound that mimics a toxic mutation, preventing growth of yeast in the presence of the inducer;

(8) screening (M7) for a compound that can mimic a

supertranscativating mutation in the p53 gene comprising:

(a) introducing a nucleic acid into the yeast cell of (M1(a)), that encodes a wildtype or a non-supertranscativating mutant p53 and comprising an inducible promoter linked to the coding sequence;

(b) plating the yeast cell and compound on raffinose medium;

(c) identifying a compound that mimics a supertranscativating mutation in p53;

(9) determining (M8) transactivation by supertransactivating p53 mutants at different expression levels and with different p53 responsive elements comprising:

(a) plating two yeast cells with two different DNA sequences that bind p53, that have been through steps (a) and (b) of (M1) on glucose, raffinose, raffinose and galactose, and raffinose and more galactose;

(b) identifying colonies on plates, where white or pink colonies indicate transactivation has occurred; and

(c) determining the level of supertransactivation of the two DNA sequences under different levels of expression by p53;

(10) detecting (M9) a supertransactivating mutation in (II) comprising:

(a) obtaining a reg1-501 mutant yeast cell of (M1(a));

(b) plating the cell on glucose and glucose with increasing concentrations of galactose; and

(c) identifying colonies on plates due to color changes;

(11) detecting (M10) a toxic mutation in (II) comprising (M9); and

(12) identifying a p53 mutant with weak transactivating activity by introducing a nucleic acid encoding p53 and comprising a promoter such as antidiuretic hormone 1 (ADH1) into a yeast cell that contains a reporter gene, plating the cell on glucose and identifying colonies due to color changes.

ACTIVITY - Cytostatic. No biological data is given.

MECHANISM OF ACTION - Gene therapy.

USE - (I) are used to induce toxicity in a cell (claimed). Mutants of p53 that are toxic, supertransactivating, or tox-suppressing are identified. (I) are used to identify compounds, agents or interactive factors, such as peptides, that mimic the toxic or supertransactivating mutations (claimed). (I) can be used to inhibit growth and treat cancer. Dwg.0/18

L10 ANSWER 2 OF 2 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1989:36809 CAPLUS

DOCUMENT NUMBER: 110:36809

TITLE: Characterization of lectins in the cotton-Verticillium dahliae Kleb. system

AUTHOR(S): Abazkhodzhaev, M. Kh.; Nuritdinova, Kh. V.; Zel'tser, S. Sh.; Adylova, A.

CORPORATE SOURCE: Inst. Eksp. Biol. Rast., Tashkent, USSR

SOURCE: Uzbekskii Biologicheskii Zhurnal (1958-199?) (1988), (4), 8-10
CODEN: UZBZAZ; ISSN: 0042-1685

DOCUMENT TYPE: Journal

LANGUAGE: Russian

AB Lectins from cotton hypocotyls inhibited conidial germination and hyphal growth in a non-virulent V. dahliae **mutant more** than in a **virulent** race. Interaction of the lectins with conidia and inductor from V. dahliae was demonstrated. Amino acid composition of 2 lectin fractions is given. In both fractions the galactose:glucose:mannose:rhamnose ratio was 4:3:1:2.

=> d his

(FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

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     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1             0 SHIGA 5A TOXIN
L2           193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3             1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4         20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5           182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6           122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7            16 L2 AND L6
L8            10 DUP REM L7 (6 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18
     NOV 2004
L9             2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
L10            2 DUP REM L9 (0 DUPLICATES REMOVED)

=> l2 and l4
L11            1 L2 AND L4

=> d ibib abs l11

L11  ANSWER 1 OF 1  WPIDS  COPYRIGHT 2004 THE THOMSON CORP on STN
ACCESSION NUMBER:     2000-412091 [35]   WPIDS
DOC. NO. CPI:         C2000-124883
TITLE:                Expression cassette used as live vector vaccine comprises
                      nucleotide sequence encoding origin of replication and
                      plasmid maintenance system which includes a
                      post-segregational killing and a partitioning function.
DERWENT CLASS:        B04 C06 D16
INVENTOR(S):          GALEN, J E
PATENT ASSIGNEE(S):   (UYMA-N) UNIV MARYLAND BALTIMORE
COUNTRY COUNT:        87
PATENT INFORMATION:

     PATENT NO        KIND DATE     WEEK      LA   PG
     ---------------------------------------------
     WO 2000032047    A1 20000608 (200035)* EN  127
        RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL
            OA PT SD SE SL SZ TZ UG ZW
         W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB
            GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU
            LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR
            TT UA UG US VN YU ZA ZW
     AU 2000020364    A  20000619 (200044)
     EP 1135025       A1 20010926 (200157)  EN
         R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
            RO SE SI
     NO 2001002721    A  20010731 (200157)
     CZ 2001001538    A3 20011114 (200175)
     HU 2001004609    A2 20020328 (200234)
     ZA 2001005383    A  20020424 (200237)      135
     US 6413768       B1 20020702 (200248)
     MX 2001005449    A1 20011201 (200282)
     JP 2003506007    W  20030218 (200315)      170
     US 6703233       B1 20040309 (200418)
     NZ 511449        A  20040528 (200437)
     US 2004161420    A1 20040819 (200455)

APPLICATION DETAILS:
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PATENT NO	KIND	APPLICATION	DATE
WO 2000032047	A1	WO 1999-US28499	19991202
AU 2000020364	A	AU 2000-20364	19991202
EP 1135025	A1	EP 1999-964042	19991202
		WO 1999-US28499	19991202
NO 2001002721	A	WO 1999-US28499	19991202
		NO 2001-2721	20010601
CZ 2001001538	A3	WO 1999-US28499	19991202
		CZ 2001-1538	19991202
HU 2001004609	A2	WO 1999-US28499	19991202
		HU 2001-4609	19991202
ZA 2001005383	A	ZA 2001-5383	20010629
US 6413768	B1	US 1998-204117	19981202
MX 2001005449	A1	MX 2001-5449	20010531
JP 2003506007	W	WO 1999-US28499	19991202
		JP 2000-584755	19991202
US 6703233	B1 CIP of Provisional	US 1998-204117	19981202
		US 1999-158738P	19991012
		US 1999-453313	19991202
NZ 511449	A	NZ 1999-511449	19991202
		WO 1999-US28499	19991202
US 2004161420	A1 CIP of Provisional Div ex	US 1998-204117	19981202
		US 1999-158738P	19991012
		US 1999-453313	19991202
		US 2004-750965	20040105

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2000020364	A Based on	WO 2000032047
EP 1135025	A1 Based on	WO 2000032047
CZ 2001001538	A3 Based on	WO 2000032047
HU 2001004609	A2 Based on	WO 2000032047
JP 2003506007	W Based on	WO 2000032047
US 6703233	B1 CIP of	US 6413768
NZ 511449	A Div in	NZ 529508
	Based on	WO 2000032047
US 2004161420	A1 CIP of	US 6413768
	Div ex	US 6703233

PRIORITY APPLN. INFO: US 1999-158738P 19991012; US 1998-204117 19981202; US 1999-453313 19991202; US 2004-750965 20040105

AN 2000-412091 [35] WPIDS

AB WO 200032047 A UPAB: 20000725

NOVELTY - An independently functioning expression cassette (I), comprises a nucleotide sequence encoding an origin of replication (ORI) and a nucleotide sequence encoding a plasmid maintenance system (PMS) which includes a post-segregational killing function (PSK) and a partitioning function.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) an amplifiable plasmid replicon (II) comprising (I);

(2) a bacterial cell comprising (II);

(3) an attenuated bacterial live vector vaccine (III), comprising a bacterial species containing a replicon comprising a nucleotide sequence encoding an antigen of interest, and a nucleotide sequence encoding a PMS;

(4) a conditionally unstable plasmid (IV), for examining changes in

plasmid stability resulting from incorporation of plasmid maintenance system, comprises an ORI yielding an average copy number of 2-75 copies and a promoter driving the expression of a protein or peptide and whose over expression imposes a metabolic burden on a bacterium, which favors plasmid loss;

(5) making (M1) a stabilized (III), which involves transforming a bacterial live vector with a replicon comprising a PMS which includes one PSK and one partitioning function, and a nucleotide sequence encoding one or more antigen;

(6) a DNA (V), comprising a modified ompC promoter phenotypically characterized so that the promoter exhibits higher rates of osmotically regulated expression in relation to a corresponding non-mutated ompC promoter; and

(7) an expression plasmid (VI) comprising (V).

ACTIVITY - Cytostatic; antibacterial; virucide; hepatropic; antiinflammatory; immunosuppressive; dermatological; antiasthmatic; antiallergic; neuroprotective; antiarthritic; antirheumatic; No supporting data is given.

MECHANISM OF ACTION - Vaccine.

USE - (IV) is used for eliciting an immune response in a human or bovine subject (claimed). (I) is used for transforming a bacterial cell which is cultured, and transformed into a subject to elicit an immune response. (I) can also be used to vaccinate a subject against Salmonella typhi. (I) may comprise an antigen for hepatitis B, Haemophilus influenzae type b, hepatitis A, acellular pertussis (acP), varicella, rotavirus, Streptococcus pneumoniae, or Neisseria meningitidis, and can be used as vaccines against diseases caused by these agents. (I) can be also used as a cancer vaccine. The antigens encoded by the plasmids are designed to provoke an immune response to autoantigens, B cell receptors and/or T cell receptors which are implicated in autoimmune or immunological diseases. Where an inappropriate immune response is raised against body tissues, or environmental antigens, the vaccines may immunize against the autoantigens, B cell receptors and/or T cell receptors to modulate the responses and ameliorate diseases, such as myasthenia gravis, lupus erythematosis, rheumatoid arthritis, multiple sclerosis, allergies and asthma.

ADVANTAGE - The plasmid maintenance systems incorporated into multicopy expression plasmids encoding one or more proteins or peptides of interest, enhances the level of expression of the protein or peptide of interest. The plasmid maintenance systems provide improved stability of recombinant plasmids, overcoming prior art problems of plasmid instability.

DESCRIPTION OF DRAWING(S) - The figure shows the pGEN expression plasmid pGEN2.

Dwg.1/8

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     (FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1            0 SHIGA 5A TOXIN
L2          193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3            1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4        20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5          182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6          122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7           16 L2 AND L6
L8           10 DUP REM L7 (6 DUPLICATES REMOVED)
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FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18 NOV 2004

L9 2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
L10 2 DUP REM L9 (0 DUPLICATES REMOVED)
L11 1 L2 AND L4

=> t ti l2 1-50

L2 ANSWER 1 OF 193 MEDLINE on STN
TI Characterization of a **shiga-toxin** 1-**resistant** stock of vero cells.

L2 ANSWER 2 OF 193 MEDLINE on STN
TI Antimicrobial resistance among enteric pathogens.

L2 ANSWER 3 OF 193 MEDLINE on STN
TI Multidrug-**resistant Shiga toxin**-producing Escherichia coli O118:H16 Latin America.

L2 ANSWER 4 OF 193 MEDLINE on STN
TI Isolation of antimicrobial-resistant Escherichia coli from retail meats purchased in Greater Washington, DC, USA.

L2 ANSWER 5 OF 193 MEDLINE on STN
TI **Shiga**-like **toxin** II impairs hepatobiliary transport of doxorubicin in rats by down-regulation of hepatic P glycoprotein and multidrug **resistance**-associated protein Mrp2.

L2 ANSWER 6 OF 193 MEDLINE on STN
TI Induction of apoptosis of human brain microvascular endothelial cells by shiga toxin 1.

L2 ANSWER 7 OF 193 MEDLINE on STN
TI Molecular basis for up-regulation by inflammatory cytokines of Shiga toxin 1 cytotoxicity and globotriaosylceramide expression.

L2 ANSWER 8 OF 193 MEDLINE on STN
TI Antibiotic **resistance**, virulence gene, and molecular profiles of **Shiga toxin**-producing Escherichia coli isolates from diverse sources in Calcutta, India.

L2 ANSWER 9 OF 193 MEDLINE on STN
TI Construction, characterization, and animal testing of WRSd1, a Shigella dysenteriae 1 vaccine.

L2 ANSWER 10 OF 193 MEDLINE on STN
TI Involvement of surface polysaccharides in the organic acid **resistance** of **Shiga Toxin**-producing Escherichia coli O157:H7.

L2 ANSWER 11 OF 193 MEDLINE on STN
TI An outbreak of diarrhoea due to multiple antimicrobial-**resistant Shiga toxin**-producing Escherichia coli O26:H11 in a nursery.

L2 ANSWER 12 OF 193 MEDLINE on STN
TI Identification and characterization of integron-mediated antibiotic **resistance** among **Shiga toxin**-producing Escherichia coli isolates.

L2 ANSWER 13 OF 193 MEDLINE on STN
TI Targeting of **Shiga toxin** B-subunit to retrograde transport route in association with detergent-**resistant** membranes.

L2 ANSWER 14 OF 193 MEDLINE on STN
TI **Resistance** patterns of non-O157 **Shiga toxin** -producing Escherichia coli (STEC) strains isolated from animals, food and asymptomatic human carriers in Switzerland.

L2 ANSWER 15 OF 193 MEDLINE on STN
TI Epidemiological analysis of Shiga toxin-producing Escherichia coli O157 isolates from familial infection.

L2 ANSWER 16 OF 193 MEDLINE on STN
TI Emergence of fosfomycin-**resistant** isolates of **Shiga** -like **toxin**-producing Escherichia coli O26.

L2 ANSWER 17 OF 193 MEDLINE on STN
TI Sequence of Shiga toxin 2 phage 933W from Escherichia coli O157:H7: Shiga toxin as a phage late-gene product.

L2 ANSWER 18 OF 193 MEDLINE on STN
TI Status of emerging drug **resistance** in **Shiga** **toxin**-producing Escherichia coli in Japan during 1996: a minireview.

L2 ANSWER 19 OF 193 MEDLINE on STN
TI Characterization of the acid **resistance** phenotype and rpoS alleles of **shiga**-like **toxin**-producing Escherichia coli.

L2 ANSWER 20 OF 193 MEDLINE on STN
TI Studies on shiga-like toxin produced by enterohemorrhagic Escherichia coli: purification and characterization of the toxin and development of methods for identifying the toxin.

L2 ANSWER 21 OF 193 MEDLINE on STN
TI Entry of Shiga toxin into cells.

L2 ANSWER 22 OF 193 MEDLINE on STN
TI Evaluation of the role of Shiga and Shiga-like toxins in mediating direct damage to human vascular endothelial cells.

L2 ANSWER 23 OF 193 MEDLINE on STN
TI Serotype, antimicrobial resistance, and adherence properties of Escherichia coli strains associated with outbreaks of diarrheal illness in children in the United States.

L2 ANSWER 24 OF 193 MEDLINE on STN
TI Pathogenesis of Shigella diarrhea. XIV. Analysis of Shiga toxin receptors on cloned HeLa cells.

L2 ANSWER 25 OF 193 MEDLINE on STN
TI Deletion of the **Shiga toxin** gene in a chlorate-**resistant** derivative of Shigella dysenteriae type 1 that retains virulence.

L2 ANSWER 26 OF 193 MEDLINE on STN
TI Isolation of **Shiga toxin-resistant** Vero cells and their use for easy identification of the toxin.

L2 ANSWER 27 OF 193 MEDLINE on STN
TI Localization of stx, a determinant essential for high-level production of shiga toxin by Shigella dysenteriae serotype 1, near pyrF and generation of stx transposon mutants.

L2 ANSWER 28 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Characterization of a **shiga**-**toxin** 1-**resistant** stock of Vero cells.

L2 ANSWER 29 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Distribution of tellurite resistance gene cluster and its resistance level in Stx-positive E. coli O157:H7.

L2 ANSWER 30 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Effectiveness of acid **resistance** mechanisms in **Shiga**-**toxin** producing Escherichia coli clones.

L2 ANSWER 31 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Multidrug-**resistant shiga toxin**-producing Escherichia coli O118:H16 in latin America.

L2 ANSWER 32 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI II Joint Meeting of the networks of the South Cone and the Amazonas for the surveillance of emergent diseases.
Original Title: II Reunion Conjunta de la red de vigilancia del Cono sur y del Amazonas para la vigilancia de las enfermedades emergentes..

L2 ANSWER 33 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Isolation of antimicrobial-resistant Escherichia coli from retail meats purchased in Greater Washington, DC, USA.

L2 ANSWER 34 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga**-like **toxin** II impairs hepatobiliary transport of doxorubicin in rats by down-regulation of hepatic P glycoprotein and multidrug **resistance**-associated protein Mrp2.

L2 ANSWER 35 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Induction of apoptosis of human brain microvascular endothelial cells by Shiga toxin 1.

L2 ANSWER 36 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Molecular basis for up-regulation by inflammatory cytokines of Shiga toxin 1 cytotoxicity and globotriaosylceramide expression.

L2 ANSWER 37 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI The surface river water and clinical Escherichia coli isolates: Characteristics, diversity and epidemiological significance.

L2 ANSWER 38 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Construction, characterization, and animal testing of WRSd1, a Shigella dysenteriae 1 vaccine.

L2 ANSWER 39 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Involvement of surface polysaccharides in the organic acid **resistance** of **Shiga toxin**-producing Escherichia coli O157:H7.

L2 ANSWER 40 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Targeting of **Shiga toxin** B-subunit to the retrograde transport route in association with detergent **resistant** membranes.

L2 ANSWER 41 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Detection and antibiotic **resistance** of **Shiga toxin** gene positive E. coli isolated from rectal swabs of children with diarrhoea in Slovakia.

L2 ANSWER 42 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Targeting of **Shiga toxin** B-subunit to the retrograde transport route in association with detergent **resistant** membranes.

L2 ANSWER 43 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI An outbreak of diarrhoea due to multiple antimicrobial-**resistant** **Shiga toxin**-producing Escherichia coli O26:H11 in a nursery.

L2 ANSWER 44 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Targeting of **Shiga toxin** B-subunit to retrograde transport route in association with detergent-**resistant** membranes.

L2 ANSWER 45 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Identification and characterization of integron-mediated antibiotic **resistance** among **Shiga toxin**-producing Escherichia coli isolates.

L2 ANSWER 46 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Resistance** patterns of non-O157 **Shiga toxin** -producing Escherichia coli (STEC) strains isolated from animals, food and asymptomatic human carriers in Switzerland.

L2 ANSWER 47 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Characterization of antimicrobial **resistance** and genetic relatedness among **Shiga-toxin** producing Escherichia coli O111 isolates.

L2 ANSWER 48 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Emergence of fosfomycin-**resistant** isolates of **Shiga** -like **toxin**-producing Escherichia coli O26.

L2 ANSWER 49 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

TI Sequence of Shiga toxin 2 phage 933W from Escherichia coli O157:H7: Shiga toxin as a phage late-gene product.

L2 ANSWER 50 OF 193 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga toxin resistance** in vero cells expressing Forssman glycolipid.

=> dup rem l2
PROCESSING COMPLETED FOR L2
L12 125 DUP REM L2 (68 DUPLICATES REMOVED)

=> t ti l12 1-50

L12 ANSWER 1 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Adaptation of Escherichia coli O157:H7 to pH alters membrane lipid composition, verotoxin secretion, and resistance to simulated gastric fluid acid

L12 ANSWER 2 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Probiotic Bifidobacteria Protect Mice from Lethal Infection with Shiga Toxin-Producing Escherichia coli O157:H7.

L12 ANSWER 3 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Characterizing spontaneous induction of Stx encoding phages using a selectable reporter system.

L12 ANSWER 4 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Refinement of a therapeutic Shiga toxin-binding probiotic for human trials.

L12 ANSWER 5 OF 125 MEDLINE on STN DUPLICATE 1
TI Characterization of a **shiga-toxin** 1-**resistant** stock of vero cells.

L12 ANSWER 6 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Non-O157 Shiga toxin-producing Escherichia coli isolated from diarrhoeic calves in Argentina

L12 ANSWER 7 OF 125 MEDLINE on STN DUPLICATE 2
TI Antimicrobial resistance among enteric pathogens.

L12 ANSWER 8 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Pentamerization of single-domain antibodies from phage libraries: A novel strategy for the rapid generation of high-avidity antibody reagents.

L12 ANSWER 9 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 3
TI Prevalence and characteristics of Shiga toxin-producing Escherichia coli, Salmonella spp. and Campylobacter spp. isolated from slaughtered sheep in Switzerland

L12 ANSWER 10 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Supervision and analysis of antibiotic **resistance** of **Shiga toxin**-producing Escherichia coli isolates

L12 ANSWER 11 OF 125 MEDLINE on STN DUPLICATE 4
TI Multidrug-**resistant Shiga toxin**-producing

Escherichia coli O118:H16 Latin America.

L12 ANSWER 12 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Escherichia coli O157:H7 Shiga toxin-encoding bacteriophages: Integrations, excisions, truncations, and evolutionary implications. [Erratum to document cited in CA139:194228]

L12 ANSWER 13 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Escherichia coli O157:H7 Shiga toxin-encoding bacteriophages: Integrations, excisions, truncations, and evolutionary implications

L12 ANSWER 14 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Immunity profiles of wild-type and recombinant Shiga-like toxin-encoding bacteriophages and characterization of novel double lysogens.

L12 ANSWER 15 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Nonpathogenic Escherichia coli can contribute to the production of Shiga toxin.

L12 ANSWER 16 OF 125 MEDLINE on STN DUPLICATE 5
TI **Shiga**-like **toxin** II impairs hepatobiliary transport of doxorubicin in rats by down-regulation of hepatic P glycoprotein and multidrug **resistance**-associated protein Mrp2.

L12 ANSWER 17 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Multidrug-**resistant shiga toxin**-producing Escherichia coli O118:H16 in Latin America [5].

L12 ANSWER 18 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 6
TI Distribution of tellurite resistance gene cluster and its resistance level in Stx-positive E. coli O157:H7.

L12 ANSWER 19 OF 125 MEDLINE on STN DUPLICATE 7
TI Isolation of antimicrobial-resistant Escherichia coli from retail meats purchased in Greater Washington, DC, USA.

L12 ANSWER 20 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Antibiotic resistance among verocytotoxigenic Escherichia coli (VTEC) and non-VTEC isolated from domestic animals and humans

L12 ANSWER 21 OF 125 MEDLINE on STN DUPLICATE 8
TI Induction of apoptosis of human brain microvascular endothelial cells by shiga toxin 1.

L12 ANSWER 22 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI (60)Co irradiation of Shiga toxin (Stx)-producing Escherichia coli induces Stx phage.

L12 ANSWER 23 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Analysis of pathogenicity islands of STEC

L12 ANSWER 24 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Antimicrobial susceptibilities of shiga toxin-producing Escherichia coli isolates from pigs with edema disease in Japan

L12 ANSWER 25 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

TI Effectiveness of acid **resistance** mechanisms in **Shiga-toxin** producing Escherichia coli clones.

L12 ANSWER 26 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI II Joint Meeting of the networks of the South Cone and the Amazonas for the surveillance of emergent diseases.
Original Title: II Reunion Conjunta de la red de vigilancia del Cono sur y del Amazonas para la vigilancia de las enfermedades emergentes..

L12 ANSWER 27 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Methods and agents for measuring and controlling multidrug resistance

L12 ANSWER 28 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 9
TI A multiresistant clone of shiga toxin-producing Escherichia coli O118:[H16] is spread in cattle and humans over different European countries

L12 ANSWER 29 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Genomic variability of O islands encoding tellurite resistance in enterohemorrhagic Escherichia coli O157:H7 isolates.

L12 ANSWER 30 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 10
TI Characterization of Shiga toxin-producing Escherichia coli O157:H7 isolated in Italy and in France

L12 ANSWER 31 OF 125 MEDLINE on STN DUPLICATE 11
TI Construction, characterization, and animal testing of WRSd1, a Shigella dysenteriae 1 vaccine.

L12 ANSWER 32 OF 125 MEDLINE on STN DUPLICATE 12
TI Antibiotic **resistance**, virulence gene, and molecular profiles of **Shiga toxin**-producing Escherichia coli isolates from diverse sources in Calcutta, India.

L12 ANSWER 33 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 13
TI Shiga-like toxin II derived from Escherichia coli O157:H7 modifies renal handling of levofloxacin in rats

L12 ANSWER 34 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Antimicrobial resistance of Escherichia coli 026, 0103, 0111, 0128, and 0145 from animals and humans.

L12 ANSWER 35 OF 125 MEDLINE on STN DUPLICATE 14
TI Molecular basis for up-regulation by inflammatory cytokines of Shiga toxin 1 cytotoxicity and globotriaosylceramide expression.

L12 ANSWER 36 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Characterization of Shiga toxin-producing Escherichia coli O26 strains and establishment of selective isolation media for these strains.

L12 ANSWER 37 OF 125 MEDLINE on STN DUPLICATE 15
TI Involvement of surface polysaccharides in the organic acid **resistance** of **Shiga Toxin**-producing Escherichia coli O157:H7.

L12 ANSWER 38 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Antimicrobial resistance of Escherichia coli O157 isolated from humans,

cattle, swine, and food.

L12 ANSWER 39 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Ferrets as a model system for renal disease secondary to intestinal infection with Escherichia coli O157:H7 and other Shiga toxin-producing E. coli.

L12 ANSWER 40 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Antimicrobial resistance of foodborne pathogens

L12 ANSWER 41 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Effect of diet on Shiga toxin-producing Escherichia coli (STEC) growth and survival in rumen and abomasum fluids.

L12 ANSWER 42 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Erratum: An outbreak of diarrhoea due to multiple antimicrobial-**resistant Shiga toxin**-producing Escherichia coli O26:H11 in a nursery (Epidemiology Infection (2001) volume 127 (221-227)).

L12 ANSWER 43 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI The surface river water and clinical Escherichia coli isolates: Characteristics, diversity and epidemiological significance.

L12 ANSWER 44 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Enhancement of pentylenetetrazole-induced seizures by Shigella dysenteriae in LPS-resistant C3H/HeJ mice: Role of the host response.

L12 ANSWER 45 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Sanitation of produce including fruits and/or vegetables involves applying bacteriophage(s) to the produce.

L12 ANSWER 46 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Performing polymerase chain reaction in a microarray of gel pads, useful for detecting bacterial toxin genes, uses immobilized primers that can be modulated selectively.

L12 ANSWER 47 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Nucleic acid sequences are used to generate universal probes and primers which can be used to identify and detect the presence of algal, archaeal, bacterial, fungal and parasitical species in a test sample.

L12 ANSWER 48 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI New recombinant microorganisms that display an oligosaccharide-comprising binding group which competes with a ligand for binding to a receptor for the ligand, used for adsorbing toxins or pathogenic organisms.

L12 ANSWER 49 OF 125 MEDLINE on STN DUPLICATE 16
TI Targeting of **Shiga toxin** B-subunit to retrograde transport route in association with detergent-**resistant** membranes.

L12 ANSWER 50 OF 125 MEDLINE on STN DUPLICATE 17
TI Identification and characterization of integron-mediated antibiotic **resistance** among **Shiga toxin**-producing Escherichia coli isolates.

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=> t ti l12 51-100

L12 ANSWER 51 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Thrombotic microangiopathy, hemolytic uremic syndrome, and thrombotic thrombocytopenic purpura.

L12 ANSWER 52 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Flavopiridol induces apoptosis and caspase-3 activation of a newly characterized Burkitt's lymphoma cell line containing mutant p53 genes.

L12 ANSWER 53 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Prevalence and characteristics of Shiga toxin-producing Escherichia coli from healthy cattle in Japan.

L12 ANSWER 54 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Practice guidelines for the management of infectious diarrhea.

L12 ANSWER 55 OF 125 MEDLINE on STN DUPLICATE 18
TI An outbreak of diarrhoea due to multiple antimicrobial-**resistant** **Shiga toxin**-producing Escherichia coli O26:H11 in a nursery.

L12 ANSWER 56 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Targeting of **Shiga toxin** B-subunit to the retrograde transport route in association with detergent **resistant** membranes.

L12 ANSWER 57 OF 125 MEDLINE on STN DUPLICATE 19
TI **Resistance** patterns of non-O157 **Shiga toxin** -producing Escherichia coli (STEC) strains isolated from animals, food and asymptomatic human carriers in Switzerland.

L12 ANSWER 58 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 20
TI Detection and antibiotic **resistance** of **Shiga** **toxin** gene positive E. coli isolated from rectal swabs of children with diarrhoea in Slovakia.

L12 ANSWER 59 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Construction of deletion mutants of Shiga (-like) toxin genes (stx-1 and/or stx-2) on enterohemorrhagic Escherichia coli (O157: H7).

L12 ANSWER 60 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Expression cassette used as live vector vaccine comprises nucleotide sequence encoding origin of replication and plasmid maintenance system which includes a post-segregational killing and a partitioning function.

L12 ANSWER 61 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Selective isolation of eae-positive strains of Shiga toxin-producing Escherichia coli.

L12 ANSWER 62 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN

TI Prevalence and characteristics of Shiga toxin-producing Escherichia coli in beef cattle slaughtered on Prince Edward Island

L12 ANSWER 63 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Iha: A novel Escherichia coli O157:H7 adherence-conferring molecule encoded on a recently acquired chromosomal island of conserved structure.

L12 ANSWER 64 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Antibacterials that are used as growth promoters in animal husbandry can affect the release of Shiga-toxin-2-converting bacteriophages and Shiga toxin 2 from Escherichia coli strains.

L12 ANSWER 65 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 21
TI PCR amplification on a microarray of gel-immobilized oligonucleotides: detection of bacterial toxin- and drug-resistant genes and their mutations

L12 ANSWER 66 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI CD40 ligand, Bcl-2, and Bcl-x(L) spare group I Burkitt lymphoma cells from CD77-directed killing via Verotoxin-1 B chain but fail to protect against the holotoxin.

L12 ANSWER 67 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Characterization of antimicrobial **resistance** and genetic relatedness among **Shiga-toxin** producing Escherichia coli O111 isolates.

L12 ANSWER 68 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Targeting of **Shiga toxin** B-subunit to the retrograde transport route in association with detergent **resistant** membranes.

L12 ANSWER 69 OF 125 MEDLINE on STN
TI Epidemiological analysis of Shiga toxin-producing Escherichia coli O157 isolates from familial infection.

L12 ANSWER 70 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Detection and characterization of Shiga toxin-producing Escherichia coli from seagulls.

L12 ANSWER 71 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Shigella and shiga toxin-producing Escherichia coli causing bloody diarrhea in Latin America.

L12 ANSWER 72 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Methods and agents for measuring and controlling multidrug resistance

L12 ANSWER 73 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI New 4-biarylbutyric and 5-biarylpentanoic acid derivatives useful as matrix metalloprotease inhibitors for treating e.g. autoimmune disease.

L12 ANSWER 74 OF 125 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Method of expressing cholera toxin B subunit protein in transgenic plants useful for producing oral vaccines.

L12 ANSWER 75 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS

RESERVED. on STN
TI Isogenic lysogens of diverse Shiga toxin 2-encoding bacteriophages produce markedly different amounts of Shiga toxin.

L12 ANSWER 76 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Comparative survival of free Shiga toxin 2-encoding phages and Escherichia coli strains outside the gut.

L12 ANSWER 77 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 22
TI Transduction of enteric Escherichia coli isolates with a derivative of shiga toxin 2-encoding bacteriophage Φ3538 isolated from Escherichia coli O157:H7

L12 ANSWER 78 OF 125 MEDLINE on STN DUPLICATE 23
TI Sequence of Shiga toxin 2 phage 933W from Escherichia coli O157:H7: Shiga toxin as a phage late-gene product.

L12 ANSWER 79 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Interaction of Shiga toxins with human brain microvascular endothelial cells: Cytokines as sensitizing agents.

L12 ANSWER 80 OF 125 MEDLINE on STN DUPLICATE 24
TI Emergence of fosfomycin-**resistant** isolates of **Shiga** -like **toxin**-producing Escherichia coli O26.

L12 ANSWER 81 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Prevalence of verotoxin-producing Escherichia coli (VTEC) in bovine coli mastitis and their antibiotic resistance patterns

L12 ANSWER 82 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Characterization of eae+ Escherichia coli isolated from healthy and diarrheic calves.

L12 ANSWER 83 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Exploiting retrograde transport of Shiga-like toxin 1 for the delivery of exogenous antigens into the MHC class I presentation pathway.

L12 ANSWER 84 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI In vivo transduction with shiga toxin 1-encoding phage.

L12 ANSWER 85 OF 125 MEDLINE on STN
TI Status of emerging drug **resistance** in **Shiga** **toxin**-producing Escherichia coli in Japan during 1996: a minireview.

L12 ANSWER 86 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Etiologies of acute, persistent, and dysenteric diarrheas in adults in Bangui, Central African Republic, in relation to human immunodeficiency virus serostatus.

L12 ANSWER 87 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Heterogeneity of phenotypic and genotypic traits including organic-acid resistance in Escherichia coli O157 isolates.

L12 ANSWER 88 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS

RESERVED. on STN
TI Detection and genetical characterization of shiga toxin-producing Escherichia coli from wild deer.

L12 ANSWER 89 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Shiga toxin-producing Escherichia coli isolates from cases of human disease show enhanced adherence to intestinal epithelial (Henle 407) cells.

L12 ANSWER 90 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga toxin resistance** via altered glycolipid expression.

L12 ANSWER 91 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga toxin resistance** via altered glycolipid expression.

L12 ANSWER 92 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga toxin resistance** in vero cells expressing Forssman glycolipid.

L12 ANSWER 93 OF 125 MEDLINE on STN DUPLICATE 25
TI Characterization of the acid **resistance** phenotype and rpoS alleles of **shiga**-like **toxin**-producing Escherichia coli.

L12 ANSWER 94 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Differentiation-associated toxin receptor modulation, cytokine production, and sensitivity to Shiga-like toxins in human monocytes and monocytic cell lines.

L12 ANSWER 95 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 26
TI Serogroups, toxins and antibiotic resistance of Escherichia coli strains isolated from diarrheic goat kids in Spain

L12 ANSWER 96 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Serogroups, toxins and antibiotic resistance of Escherichia coli strains isolated from diarrheic lambs in Spain

L12 ANSWER 97 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Shigella infections.

L12 ANSWER 98 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI DNA fingerprinting of pathogenic bacteria by fluorophore-enhanced repetitive sequence-based polymerase chain reaction.

L12 ANSWER 99 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Evidence that proteolytic separation of Shiga-like toxin type IIv A subunit into A1 and A2 subunits is not required for toxin activity.

L12 ANSWER 100 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Intracellular expression of toxic shock syndrome toxin 1 in Saccharomyces cerevisiae.

=> t ti l12 101-125

L12 ANSWER 101 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Pathogenesis of Shigella diarrhea: XVII. A mammalian cell membrane glycolipid, Gb3, is required but not sufficient to confer sensitivity to Shiga toxin.

L12 ANSWER 102 OF 125 MEDLINE on STN DUPLICATE 27
TI Entry of Shiga toxin into cells.

L12 ANSWER 103 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Differential effects of globotriaosylceramide (Gb3) and globotetraosylceramide (Gb4) treatment of toxin **resistant** cells on their interaction with **Shiga toxin** (ShT) and **Shiga**-like **toxin** II pig variant (SLTIIvp).

L12 ANSWER 104 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Distribution and characteristics of verocytotoxigenic Escherichia coli isolated from Ontario dairy cattle.

L12 ANSWER 105 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Diarrhoeal disease in children less than one year of age at a children's hospital in Guangzhou, People's Republic of China.

L12 ANSWER 106 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI Verotoxin-resistant cell clones are deficient in the glycolipid globotriosylceramide: differential basis of phenotype

L12 ANSWER 107 OF 125 MEDLINE on STN DUPLICATE 28
TI Evaluation of the role of Shiga and Shiga-like toxins in mediating direct damage to human vascular endothelial cells.

L12 ANSWER 108 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Laboratory investigations on the low pathogenic potential of Plesiomonas shigelloides.

L12 ANSWER 109 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Shiga-like toxin-producing E. coli infections in Korea.

L12 ANSWER 110 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Prevalence of verocytotoxigenic Escherichia coli in ground beef, pork, and chicken in southwestern Ontario.

L12 ANSWER 111 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Expression of glycolipid receptors to Shiga-like toxin on human B lymphocytes: a mechanism for the failure of long-lived antibody response to dysenteric disease.

L12 ANSWER 112 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Bacterial endotoxin both enhances and inhibits the toxicity of Shiga-like toxin II in rabbits and mice.

L12 ANSWER 113 OF 125 MEDLINE on STN DUPLICATE 29
TI Serotype, antimicrobial resistance, and adherence properties of Escherichia coli strains associated with outbreaks of diarrheal illness in children in the United States.

L12 ANSWER 114 OF 125 MEDLINE on STN DUPLICATE 30
TI Pathogenesis of Shigella diarrhea. XIV. Analysis of Shiga toxin receptors on cloned HeLa cells.

L12 ANSWER 115 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI ANALYSIS OF SHIGELLA-DYSENTERIAE TYPE 1 STRAINS FROM A RECENT OUTBREAK IN CENTRAL AMERICA BY STANDARD AND MOLECULAR EPIDEMIOLOGIC TECHNIQUES.

L12 ANSWER 116 OF 125 MEDLINE on STN
TI Studies on shiga-like toxin produced by enterohemorrhagic Escherichia coli: purification and characterization of the toxin and development of methods for identifying the toxin.

L12 ANSWER 117 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI ESCHERICHIA-COLI STRAINS ISOLATED DURING OUTBREAKS OF DIARRHEAL ILLNESS IN CHILDREN IN THE USA.

L12 ANSWER 118 OF 125 MEDLINE on STN DUPLICATE 31
TI Isolation of **Shiga toxin-resistant** Vero cells and their use for easy identification of the toxin.

L12 ANSWER 119 OF 125 MEDLINE on STN DUPLICATE 32
TI Deletion of the **Shiga toxin** gene in a chlorate-**resistant** derivative of Shigella dysenteriae type 1 that retains virulence.

L12 ANSWER 120 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Analysis and genetic manipulation of Shigella virulence determinants for vaccine development.

L12 ANSWER 121 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Production of a unique cytotoxin by Campylobacter jejuni.

L12 ANSWER 122 OF 125 MEDLINE on STN DUPLICATE 33
TI Localization of stx, a determinant essential for high-level production of shiga toxin by Shigella dysenteriae serotype 1, near pyrF and generation of stx transposon mutants.

L12 ANSWER 123 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI The relation between dysentery and the autonomic nervous system. I. Research on rabbits

L12 ANSWER 124 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI An experimental study on the relation between dysentery and vitamin C. II. The influence of vitamin C on the infection of rats with Ohara (Kruse-Sonne) bacilli; and the **resistance** of rabbits against **Shiga** [bacillus] **toxin** mixed with vitamin C

L12 ANSWER 125 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
TI An experimental study on the relation between dysentery and vitamin C. II. The influence of vitamin C on the infection of rats with Ohara (Kruse-Sonne) bacilli; and the **resistance** of rabbits against

Shiga [bacillus] **toxin** mixed with vitamin C

=> d ibib abs L12 4,15,49,90,92,94,101-103, 106-108, 111-114, 118-125

L12 ANSWER 4 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 2004220273 EMBASE
TITLE: Refinement of a therapeutic Shiga toxin-binding probiotic for human trials.
AUTHOR: Pinyon R.A.; Paton J.C.; Paton A.W.; Bottan J.A.; Morona R.
CORPORATE SOURCE: Prof. J.C. Paton, Sch. of Molec. and Biomed. Science, University of Adelaide, Adelaide, SA 5005, Australia. james.paton@adelaide.edu.au
SOURCE: Journal of Infectious Diseases, (1 May 2004) 189/9 (1547-1555).
Refs: 33
ISSN: 0022-1899 CODEN: JIDIAQ
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB We have previously constructed a recombinant bacterium expressing a modified lipopolysaccharide (LPS) mimicking the **Shiga toxin** receptor, which binds **toxin** with high avidity. This involved cloning Neisseria galactosyl transferase genes (IgtC and IgtE) in pK184 in a derivative of Escherichia coli R1 (CWG308). Such constructs have considerable potential for prevention of disease caused by **Shiga toxin**-producing E. coli (STEC). However, neither the E. coli host strain nor the expression plasmid is suitable for human use, because the former is derived from a clinical isolate and the latter contains a kanamycin-**resistance** gene. We have constructed, as a prelude to human trials, a nonpathogenic E. coli K-12 C600 derivative with deletions in waaO and waaB, such that it has the same LPS core structure as CWG308. We also deleted the thyA gene from this strain, rendering it thymine dependent. The kanamycin-**resistance** gene was also deleted from pK184 and was replaced with Salmonella typhimurium thyA. Neisseria IgtCE was then cloned into this plasmid and transformed into C600ΔwaaOBΔthyA. The plasmid was stably maintained, and the construct produced a modified LPS and neutralized Stx1 and Stx2c. Moreover, mice challenged with an otherwise fatal dose of STEC were completely protected by oral administration of the novel construct.

L12 ANSWER 15 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 2003219673 EMBASE
TITLE: Nonpathogenic Escherichia coli can contribute to the production of Shiga toxin.
AUTHOR: Gamage S.D.; Strasser J.E.; Chalk C.L.; Weiss A.A.
CORPORATE SOURCE: A.A. Weiss, Department of Molecular Genetics, University of Cincinnati, 231 Albert Sabin Way, Cincinnati, OH 45267-0524, United States. Alison.Weiss@uc.edu
SOURCE: Infection and Immunity, (1 Jun 2003) 71/6 (3107-3115).
Refs: 41
ISSN: 0019-9567 CODEN: INFIBR
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
037 Drug Literature Index

048 Gastroenterology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The food-borne pathogen, Escherichia coli O157:H7, has been associated with gastrointestinal disease and the life-threatening sequela hemolytic uremic syndrome. The genes for the virulence factor, **Shiga toxin** 2 (Stx2), in E. coli O157:H7 are encoded on a temperate bacteriophage under the regulation of the late gene promoter. Induction of the phage lytic cycle is required for toxin synthesis and release. We investigated the hypothesis that nonpathogenic E. coli could amplify Stx2 production if infected with the toxin-encoding phage. Toxin-encoding phage were incubated with E. coli that were either susceptible or **resistant** to the phage. The addition of phage to phage-susceptible bacteria resulted in up to 40-fold more toxin than a pure culture of lysogens, whereas the addition of phage to phage-**resistant** bacteria resulted in significantly reduced levels of toxin. Intestinal E. coli isolates incubated with **Shiga toxin**-encoding phage produced variable amounts of toxin. Of 37 isolates, 3 produced significantly more toxin than was present in the inoculum, and 1 fecal isolate appeared to inactivate the toxin. Toxin production in the intestine was assessed in a murine model. Fecal toxin recovery was significantly reduced when phage-**resistant** E. coli was present. These results suggest that the susceptibility of the intestinal flora to the **Shiga toxin** phage could exert either a protective or an antagonistic influence on the severity of disease by pathogens with phage-encoded **Shiga toxin**. **Toxin** production by intestinal flora may represent a novel strategy of pathogenesis.

L12 ANSWER 49 OF 125 MEDLINE on STN DUPLICATE 16
ACCESSION NUMBER: 2001469748 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11514628
TITLE: Targeting of **Shiga toxin** B-subunit to retrograde transport route in association with detergent-**resistant** membranes.
AUTHOR: Falguieres T; Mallard F; Baron C; Hanau D; Lingwood C; Goud B; Salamero J; Johannes L
CORPORATE SOURCE: Unite Mixte de Recherche 144 Institut Curie/ Centre National de la Recherche Scientifique, F-75248 Paris Cedex 05, France.
SOURCE: Molecular biology of the cell, (2001 Aug) 12 (8) 2453-68. Journal code: 9201390. ISSN: 1059-1524.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200201
ENTRY DATE: Entered STN: 20010830
Last Updated on STN: 20020201
Entered Medline: 20020131
AB In HeLa cells, Shiga toxin B-subunit is transported from the plasma membrane to the endoplasmic reticulum, via early endosomes and the Golgi apparatus, circumventing the late endocytic pathway. We describe here that in cells derived from human monocytes, i.e., macrophages and dendritic cells, the B-subunit was internalized in a receptor-dependent manner, but retrograde transport to the biosynthetic/secretory pathway did not occur and part of the internalized protein was degraded in lysosomes. These differences correlated with the observation that the B-subunit associated with Triton X-100-resistant membranes in HeLa cells, but not in monocyte-derived cells, suggesting that retrograde targeting to the biosynthetic/secretory pathway required association with specialized microdomains of biological membranes. In agreement with this hypothesis we found that in HeLa cells, the B-subunit resisted extraction by Triton

X-100 until its arrival in the target compartments of the retrograde pathway, i.e., the Golgi apparatus and the endoplasmic reticulum. Furthermore, destabilization of Triton X-100-resistant membranes by cholesterol extraction potently inhibited B-subunit transport from early endosomes to the trans-Golgi network, whereas under the same conditions, recycling of transferrin was not affected. Our data thus provide first evidence for a role of lipid asymmetry in membrane sorting at the interface between early endosomes and the trans-Golgi network.

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ACCESSION NUMBER: 1997:473971 BIOSIS
DOCUMENT NUMBER: PREV199799773174
TITLE: **Shiga toxin resistance** via altered glycolipid expression.
AUTHOR(S): Elliott, Sean P.; Haslam, David B.
CORPORATE SOURCE: Dep. Pediatr., Washington Univ. Sch. Med., St. Louis, MO, USA
SOURCE: Clinical Infectious Diseases, (1997) Vol. 25, No. 2, pp. 426.
Meeting Info.: 35th Annual Meeting of the Infectious Diseases Society of America.
CODEN: CIDIEL. ISSN: 1058-4838.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 4 Nov 1997
Last Updated on STN: 4 Nov 1997

L12 ANSWER 92 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

ACCESSION NUMBER: 1998:19897 BIOSIS
DOCUMENT NUMBER: PREV199800019897
TITLE: **Shiga toxin resistance** in vero cells expressing Forssman glycolipid.
AUTHOR(S): Haslam, David; Elliott, Sean P.
CORPORATE SOURCE: Dep. Pediatr., Washington Univ. Sch. Med., St. Louis, MO, USA
SOURCE: Molecular Biology of the Cell, (Nov., 1997) Vol. 8, No. SUPPL., pp. 80A. print.
Meeting Info.: 37th Annual Meeting of the American Society for Cell Biology. Washington, D.C., USA. December 13-17, 1997. American Society for Cell Biology.
CODEN: MBCEEV. ISSN: 1059-1524.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
Conference; (Meeting Poster)
LANGUAGE: English
ENTRY DATE: Entered STN: 5 Jan 1998
Last Updated on STN: 5 Jan 1998

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ACCESSION NUMBER: 96100232 EMBASE
DOCUMENT NUMBER: 1996100232
TITLE: Differentiation-associated toxin receptor modulation, cytokine production, and sensitivity to Shiga-like toxins in human monocytes and monocytic cell lines.
AUTHOR: Ramegowda B.; Tesh V.L.
CORPORATE SOURCE: Med. Microbiology/Immunology Dept., Texas A/M Univ. Health Science Ctr.,College Station, TX 77843-1114, United States
SOURCE: Infection and Immunity, (1996) 64/4 (1173-1180).

ISSN: 0019-9567 CODEN: INFIBR
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Infections with **Shiga toxin**-producing Shigella dysenteriae type 1 or **Shiga**-like **toxin** (SLT)-producing Escherichia coli cause bloody diarrhea and are associated with an increased risk of acute renal failure and severe neurological complications. Histopathological examination of human and animal tissues suggests that the target cells for toxin action are vascular endothelial cells. Proinflammatory cytokines regulate endothelial cell membrane expression of the glycolipid globotriaosylceramide (Gb3) which serves as the toxin receptor, suggesting that the host response to the toxins or other bacterial products may contribute to pathogenesis by regulating target cell sensitivity to the toxins. We examined the effects of purified SLTs on human peripheral blood monocytes (PBMn) and two monocytic cell lines. Undifferentiated THP-1 cells were sensitive to SLTs. Treatment of the cells with a number of differentiation factors resulted in increased toxin **resistance** which was associated with decreased toxin receptor expression. U- 937 cells, irrespective of maturation state, and PBMn were **resistant** to the toxins. U-937 cells expressed low levels of Gb3, and toxin receptor expression was not altered during differentiation. Treatment of monocytic cells with tumor necrosis factor alpha (TNF-α) did not markedly increase sensitivity or alter toxin receptor expression. Undifferentiated monocytic cells failed to synthesize TNF and interleukin 1β when treated with sublethal concentrations of SLT type I (SLT-I), whereas cells treated with 12-O-tetradecanoylphorbol-13-acetate acquired the ability to produce cytokines when stimulated with SLT-I. When stimulated with SLT-I, U-937 cells produced lower levels of TNF than PBMn and THP-I cells did.

L12 ANSWER 101 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 94070915 EMBASE
DOCUMENT NUMBER: 1994070915
TITLE: Pathogenesis of Shigella diarrhea: XVII. A mammalian cell membrane glycolipid, Gb3, is required but not sufficient to confer sensitivity to Shiga toxin.
AUTHOR: Jacewicz M.S.; Mobassaleh M.; Gross S.K.; Balasubramanian K.A.; Daniel P.F.; Raghavan S.; McCluer R.H.; Keusch G.T.
CORPORATE SOURCE: Geographic Med./Infectious Dis. Div., New England Medical Center, Box 041, 750 Washington St.,Boston, MA 02111, United States
SOURCE: Journal of Infectious Diseases, (1994) 169/3 (538-546). ISSN: 0022-1899 CODEN: JIDIAQ
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
048 Gastroenterology
052 Toxicology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB **Shiga toxin** recognizes a galactose-α1→4-galactose terminal glycolipid, globotriaosylceramide (Gb3), in sensitive mammalian cells and is translocated by endocytosis to the cytoplasm, where it blocks protein synthesis. To determine if Gb3 is both required and sufficient for toxicity, Gb3 content in cells was altered by blocking key biosynthetic or degradative path enzymes with specific inhibitors. The resulting decrease or increase in cellular Gb3 was associated with a

decrease or increase in binding of and response to **Shiga toxin**. **Toxin-resistant** Gb3-deficient variants of sensitive cells fused with liposomes containing Gb3 but not globotetraosylceramide (Gb4) became susceptible, whereas fusion of Gb3 liposomes to naturally **resistant** Gb3- deficient CHO cells increased toxin binding but not cytotoxicity. These data demonstrate that Gb3 is required, but not sufficient, for the action of **Shiga toxin** and suggest the existence of a toxin translocation mechanism linked to surface glycolipids that is not expressed in CHO cells.

L12 ANSWER 102 OF 125 MEDLINE on STN DUPLICATE 27
ACCESSION NUMBER: 93350341 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8347933
TITLE: Entry of Shiga toxin into cells.
AUTHOR: Sandvig K; Dubinina E; Garred O; Prydz K; Kozlov J V; Hansen S H; Van Deurs B
CORPORATE SOURCE: Institute for Cancer Research, Norwegian Radium Hospital, Montebello, Oslo.
SOURCE: Zentralblatt fur Bakteriologie : international journal of medical microbiology, (1993 Apr) 278 (2-3) 296-305. Journal code: 9203851. ISSN: 0934-8840.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199309
ENTRY DATE: Entered STN: 19931001
Last Updated on STN: 20000303
Entered Medline: 19930916

AB The effect of Shiga toxin with mutations in the A fragment has been tested on cells in order to get more information about the processing of the A fragment during entry into the cytosol. A mutant with a deletion between the A1 and A2 domain in the A fragment is resistant to cleavage by trypsin and is less toxic than wild type toxin on both Vero and A431 cells. The results support the view that processing of the A fragment is important for the high toxicity of the wild type toxin. A number of cell lines are **resistant** to **Shiga toxin** although they bind the **toxin**. However, A431 cells can be sensitized by butyric acid treatment, and transport of Shiga toxin to the Golgi apparatus seems to be required for the intoxication in the sensitized cells. The role of retrograde transport through the Golgi apparatus to the endoplasmic reticulum (ER) will be discussed.

L12 ANSWER 103 OF 125 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 1993:357047 BIOSIS
DOCUMENT NUMBER: PREV199345040472
TITLE: Differential effects of globotriaosylceramide (Gb3) and globotetraosylceramide (Gb4) treatment of toxin **resistant** cells on their interaction with **Shiga toxin** (ShT) and **Shiga**-like **toxin** II pig variant (SLTIIvp).
AUTHOR(S): Jacewicz, M. [Reprint author]; Acheson, D.; Donohue-Rolfe, A.; Kane, A.; Keusch, T. G.
CORPORATE SOURCE: New England Med. Center, Boston, MA, USA
SOURCE: Abstracts of the General Meeting of the American Society for Microbiology, (1993) Vol. 93, No. 0, pp. 41. Meeting Info.: 93rd General Meeting of the American Society for Microbiology. Atlanta, Georgia, USA. May 16-20, 1993. ISSN: 1060-2011.
DOCUMENT TYPE: Conference; (Meeting)
LANGUAGE: English

ENTRY DATE: Entered STN: 31 Jul 1993
Last Updated on STN: 31 Aug 1993

L12 ANSWER 106 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:183099 CAPLUS
DOCUMENT NUMBER: 114:183099
TITLE: Verotoxin-resistant cell clones are deficient in the glycolipid globotriosylceramide: differential basis of phenotype
AUTHOR(S): Pudymaitis, A.; Armstrong, G.; Lingwood, C. A.
CORPORATE SOURCE: Res. Inst., Hosp. Sick Child., Toronto, ON, M5G 1X8, Can.
SOURCE: Archives of Biochemistry and Biophysics (1991), 286(2), 448-52
CODEN: ABBIA4; ISSN: 0003-9861
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Escherichia coli-derived verotoxin is a toxic protein highly selective toward certain primate cells. Two susceptible cell lines are the Daudi cell line (human Burkitt lymphoma) and the Vero cell line (Green African monkey kidney). These cell lines contain significant levels of the verotoxin-binding glycolipid globotriosylceramide (Gb3). A clone was selected from the Vero cell line for resistance to Verotoxin 2, while a mutant from the Daudi cell line was selected for resistance to Verotoxin 1. Both were deficient in globotriosylceramide with a corresponding increase in the precursor glycolipid lactosylceramide. Cell free assay of α-galactosyltransferase activity revealed that the Vero cell clone (VRP) contained reduced enzyme activity, whereas in the Daudi mutant (VT20) no decrease was noted in vitro. The observations suggest a complex regulation of Gb3 biosynthesis which is related to P blood group antigen expression.

L12 ANSWER 107 OF 125 MEDLINE on STN DUPLICATE 28
ACCESSION NUMBER: 91311099 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1649877
TITLE: Evaluation of the role of Shiga and Shiga-like toxins in mediating direct damage to human vascular endothelial cells.
AUTHOR: Tesh V L; Samuel J E; Perera L P; Sharefkin J B; O'Brien A D
CORPORATE SOURCE: Department of Microbiology, Uniformed Services University of the Health Sciences, Bethesda, Maryland 20814.
CONTRACT NUMBER: AI-20148 (NIAID)
SOURCE: Journal of infectious diseases, (1991 Aug) 164 (2) 344-52.
Journal code: 0413675. ISSN: 0022-1899.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199108
ENTRY DATE: Entered STN: 19910913
Last Updated on STN: 19970203
Entered Medline: 19910823
AB Infection with Shiga toxin- and Shiga-like toxin-producing strains of Shigella dysenteriae and Escherichia coli, respectively, can progress to the hemolytic-uremic syndrome. It has been hypothesized that circulating Shiga toxin, Shiga-like toxins, and endotoxins may contribute to the disease by directly damaging glomerular endothelial cells. The effects of these toxins on HeLa, Vero, and human vascular endothelial cells (EC) were examined. Confluent EC were sensitive to Shiga toxin but were at least 10(6)-fold less sensitive to the toxins than were Vero cells. Shiga toxin was the predominant cytotoxic factor. Lipopolysaccharides were not

cytotoxic and did not augment Shiga toxin-mediated toxicity. Lower doses of Shiga toxin caused cytotoxicity when coincubated with tumor necrosis factor. The relative **resistance** of EC to **Shiga toxin** and **Shiga**-like toxins may be due to reduced toxin binding, as low levels of globotriaosylceramide (Gb3), the toxin-specific receptor, were found in EC membranes.

L12 ANSWER 108 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 91039119 EMBASE
DOCUMENT NUMBER: 1991039119
TITLE: Laboratory investigations on the low pathogenic potential of Plesiomonas shigelloides.
AUTHOR: Abbott S.L.; Kokka R.P.; Janda J.M.
CORPORATE SOURCE: Microbial Diseases Laboratory, California Department of, Health Services,Berkeley, CA 94704, United States
SOURCE: Journal of Clinical Microbiology, (1991) 29/1 (148-153). ISSN: 0095-1137 CODEN: JCMIDW
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The pathogenic properties of 16 Plesiomonas shigelloides strains recovered from humans with extraintestinal and intestinal illnesses, infected animals, and environmental sources were investigated. Most strains possessed a high cell charge and low surface hydrophobicity analogous to those of Shigella spp.; additionally, serogroup O:17 strains reacted with Shigella group D antisera. However, unlike the shigellae, P. shigelloides strains did not universally bind Congo red, were noninvasive in HEp-2 cell assays, and did not produce a **Shiga**-like **toxin** on Vero cells. On HEp-2, Y1, and possibly Vero cells, a low-level cytolysin was consistently produced by all 16 P. shigelloides strains when grown in either Evan Casamino Acids-yeast extract or Penassay broth. The median 50% lethal dose for all 16 P. shigelloides strains in outbred Swiss Webster mice was 3.5 x 108 CFU (range, 3.2 x 107 to > 1 x 109 CFU). Animal pathogenicity did not correlate with cytolysin expression, possession of a ≥ 120-MDa plasmid, protein profile, or **resistance** to complement-mediated lysis. No strain analyzed produced siderophores or a heat-stable enterotoxin. The results suggest that members of the genus Plesiomonas have an overall low pathogenic potential, irrespective of the site of isolation or phenotypic, serologic, or surface properties shared with other traditional enteropathogens.

L12 ANSWER 111 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 90041190 EMBASE
DOCUMENT NUMBER: 1990041190
TITLE: Expression of glycolipid receptors to Shiga-like toxin on human B lymphocytes: a mechanism for the failure of long-lived antibody response to dysenteric disease.
AUTHOR: Cohen A.; Madrid-Marina V.; Estrov Z.; Freedman M.H.; Lingwood C.A.; Dosch H.M.
CORPORATE SOURCE: Div. Immunology/Rheumatology, Research Institute, The Hospital for Sick Children, 555 University Avenue,Toronto, Ont., Canada
SOURCE: International Immunology, (1990) 2/1 (1-8). ISSN: 0953-8178 CODEN: INIMEN
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
025 Hematology

026 Immunology, Serology and Transplantation
052 Toxicology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Fresh and transformed human B lineage cells were found to be sensitive to the cytotoxic action of **Shiga**-like **toxin** (SLT), a bacterial cytotoxin. The toxin was specifically bound by the glycolipids globotriosylceramide and galabiosylceramide expressed on the surface of sensitive cells. Mutant Daudi cells selected for **resistance** to SLT cytotoxicity (SLT20(R)) were deficient in SLT-binding glycolipids and failed to bind SLT to their surface, suggesting a role for these glycolipids in the mediation of SLT cytotoxicity. Of a number of normal and transformed lymphoid and myeloid cells screened for SLT sensitivity, only B lymphoid cells were susceptible to SLT action. Moreover, B lymphoid cells were the only cells expressing the SLT binding glycolipids. In vitro B cell activation studies with Epstein-Barr virus and pokeweed mitogen both indicated that the vast majority of SLT-sensitive B cells belong to the IgG and IgA committed subset, whereas most IgM and IgM/D producing cells were **resistant** to SLT toxicity. The selective elimination of IgG and IgA committed cells may explain the production of only lgM class anti-SLT antibodies in Shigella-infected humans leading to the failure of long-term immunity to dysenteric disease.

L12 ANSWER 112 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 89259919 EMBASE
DOCUMENT NUMBER: 1989259919
TITLE: Bacterial endotoxin both enhances and inhibits the toxicity of Shiga-like toxin II in rabbits and mice.
AUTHOR: Barrett T.J.; Potter M.E.; Wachsmuth I.K.
CORPORATE SOURCE: Division of Bacterial Diseases, Center for Infectious Diseases, Centers for Disease Control, Atlanta, GA 30333, United States
SOURCE: Infection and Immunity, (1989) 57/11 (3434-3437).
ISSN: 0019-9567 CODEN: INFIBR
COUNTRY: United States
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
048 Gastroenterology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The ability of bacterial lipopolysaccharide (LPS) to enhance the toxicity of **Shiga**-like **toxin** II (SLT-II) was investigated in rabbits and mice. Rabbits were continuously infused with 0.5 50% lethal dose (LD50) of SLT-II per day. Rabbits that received a 30-μg/kg dose of LPS (0.02 LD50) on day 3 of infusion were significantly more likely to die than were rabbits receiving SLT-II only. Rabbits receiving SLT-II and a lower dose of LPS (3 μg/kg) did not die but lost an average 3.3% ± 1.0% of initial body weight during the first 5 days of infusion, compared with weight gains of 4.2% ± 0.6% and 17.1% ± 0.9% for rabbits receiving only SLT-II or LPS, respectively. Rabbits that were pretreated with LPS 20 h before challenge with a single dose of SLT-II showed highly significant protection from both the diarrheagenic and lethal effects of SLT-II. Pretreatment of endotoxin-responsive C3H/HeN mice protected the animals from challenge with an LD50 but not an LD100 of SLT-II. LPS enhanced the lethal toxicity of SLT-II for C3H/HeN mice when it was given at 8 or 24 h but not 0 or 72 h after SLT-II challenge. LPS did not affect the lethal toxicity of SLT-II for endotoxin-**resistant** C3H/HeJ mice. These results suggest that LPS enhances the effects of SLT-II in vivo. Since cecal changes that increase mucosal permeability occur in response to SLT in rabbits, this synergy may be directly relevant to disease processes.

L12 ANSWER 113 OF 125 MEDLINE on STN DUPLICATE 29
ACCESSION NUMBER: 90062470 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2685024
TITLE: Serotype, antimicrobial resistance, and adherence properties of Escherichia coli strains associated with outbreaks of diarrheal illness in children in the United States.
AUTHOR: Moyenuddin M; Wachsmuth I K; Moseley S L; Bopp C A; Blake P A
CORPORATE SOURCE: Enteric Diseases Branch, Centers for Disease Control, Atlanta, Georgia 30333.
SOURCE: Journal of clinical microbiology, (1989 Oct) 27 (10) 2234-9.
Journal code: 7505564. ISSN: 0095-1137.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199001
ENTRY DATE: Entered STN: 19900328
Last Updated on STN: 19900328
Entered Medline: 19900109

AB Since most recorded outbreaks of diarrhea in U.S. infants attributed to Escherichia coli occurred before currently available pathogenicity assays existed, we examined the characteristics of nonenterotoxigenic E. coli strains isolated from 50 outbreaks of diarrheal disease in U.S. infants between 1934 and 1987. We assayed the strains for enteropathogenic E. coli (EPEC) serotype, localized adherence (LA) and diffuse adherence to tissue cultures, the presence of EPEC adherence factor genes, **Shiga**-like (Vero) **toxin** production, and antimicrobial **resistance**. EPEC serotypes were identified in 28 outbreaks (56%). LA to HeLa cells was found in 23 outbreak strains and correlated 100% with the EPEC adherence factor probe. LA was observed in 21 of 28 EPEC and 2 of 22 non-EPEC strains; however, 5 of 23 strains that were LA positive for HeLa cells did not adhere to HEp-2 or HL cells. One strain was diffuse adherence positive, and none was Shiga-like toxin positive. Multiple resistance was common in EPEC (64%), LA-positive (74%), and LA-positive EPEC (76%) strains but not in others (10%). EPEC serotypes or LA was found in 60% (n = 30) of the outbreak strains. The remaining E. coli strains may represent nonpathogenic normal flora, as-yet-undefined pathogens, or pathogens that have lost virulence-associated traits during storage or subculturing.

L12 ANSWER 114 OF 125 MEDLINE on STN DUPLICATE 30
ACCESSION NUMBER: 89215359 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2651533
TITLE: Pathogenesis of Shigella diarrhea. XIV. Analysis of Shiga toxin receptors on cloned HeLa cells.
AUTHOR: Jacewicz M; Feldman H A; Donohue-Rolfe A; Balasubramanian K A; Keusch G T
CORPORATE SOURCE: Division of Geographic Medicine and Infectious Diseases, New England Medical Center, Boston, MA 02111.
CONTRACT NUMBER: AI-16242 (NIAID)
AI-20235 (NIAID)
DK-34928 (NIDDK)
SOURCE: Journal of infectious diseases, (1989 May) 159 (5) 881-9.
Journal code: 0413675. ISSN: 0022-1899.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals

ENTRY MONTH: 198905
ENTRY DATE: Entered STN: 19900306
Last Updated on STN: 19970203
Entered Medline: 19890526

AB Binding kinetics of Shiga toxin to HeLa CCL-2 cells and to cell lines cloned by limiting dilutions were determined. Lines with a wide range of sensitivity to Shiga toxin were obtained. Binding data, analyzed by a computer-based Scatchard model program, revealed two classes of binding sites, one of low affinity and high capacity and one of high affinity and low capacity. The number of high affinity, but not low affinity, sites present on the clones correlated with their sensitivity to toxin. Tunicamycin-treated CCL-2 cells became **resistant** to **Shiga toxin** in parallel with a reduction in the capacity of the high-affinity site. Cell content of Gb3, the glycolipid receptor for Shiga toxin, decreased as the sensitivity of the cells diminished. These data show that a minority of Shiga toxin binding sites of HeLa cells are involved in the cytotoxic response and suggest that Gb3 is the high-affinity functional cytotoxin receptor.

L12 ANSWER 118 OF 125 MEDLINE on STN DUPLICATE 31
ACCESSION NUMBER: 88314296 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3045003
TITLE: Isolation of **Shiga toxin-resistant** Vero cells and their use for easy identification of the toxin.
AUTHOR: Kongmuang U; Honda T; Miwatani T
CORPORATE SOURCE: Research Institute for Microbial Diseases, Osaka University, Japan.
SOURCE: Infection and immunity, (1988 Sep) 56 (9) 2491-4.
Journal code: 0246127. ISSN: 0019-9567.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198810
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19900308
Entered Medline: 19881004

AB **Shiga toxin-resistant** Vero cells were isolated by treatment of the cells with nitrosoguanidine. These mutant cells were not affected by Shiga toxin at more than 1 microgram/ml, although the parent Vero cells were sensitive to 25 pg of the toxin per ml. Immunofluorescence studies showed that all the mutant cells had lost toxin-binding capacity. The cytotoxic activities of various bacterial cultures against the parent and mutant cells were compared. All samples from 10 strains of Shigella dysenteriae type 1 and all three strains of Escherichia coli O157:H7 tested showed cytotoxicity to the parent cells but not to the mutant cells. Samples from other organisms, such as Shigella flexneri, Shigella sonnei, Clostridium difficile, Aeromonas hydrophila, Aeromonas sobria, and other E. coli strains, either had no effect or were cytotoxic on both the parent and mutant cells. Thus, these mutant cells could be used to identify Shiga-like toxin and distinguish it from other cytotoxins. The results also suggest the presence of a receptor for Shiga-like toxin on Vero cells that is essential for expression of the cytotoxicity of Shiga toxin but is not essential for growth of Vero cells.

L12 ANSWER 119 OF 125 MEDLINE on STN DUPLICATE 32
ACCESSION NUMBER: 89009978 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3049838
TITLE: Deletion of the **Shiga toxin** gene in a chlorate-**resistant** derivative of Shigella

dysenteriae type 1 that retains virulence.
AUTHOR: Neill R J; Gemski P; Formal S B; Newland J W
CORPORATE SOURCE: Department of Biological Chemistry, Walter Reed Army Institute of Research, Washington, D.C. 20307.
SOURCE: Journal of infectious diseases, (1988 Oct) 158 (4) 737-41. Journal code: 0413675. ISSN: 0022-1899.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 198811
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19970203
Entered Medline: 19881109

AB We used a probe specific for detecting the structural-gene sequences of Shiga toxin to analyze the genetic nature of toxin synthesis in mutant derivatives of Shigella dysenteriae type 1. A chlorate-**resistant** (chl) mutant (725-78) of S. dysenteriae type 1 strain 3818T, which had retained virulence but had lost production of high levels of cytotoxic activity associated with **Shiga toxin** synthesis, contained a complete deletion of the **Shiga toxin** structural-gene sequences. These structural-gene sequences were also absent in a derivative of S. dysenteriae type 1 that contained a substitution of Escherichia coli DNA in the trp region of the chromosome. Isolates of Shigella flexneri and Shigella sonnei also did not react with the probe. The low-level cytotoxic activities associated with the mutant S. dysenteriae type 1 strains or with the virulent S. flexneri and S. sonnei strains are neutralizable with antiserum to Shiga toxin; however, these cytotoxic activities are not determined by the genes encoding classic Shiga toxin.

L12 ANSWER 120 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 88089133 EMBASE
DOCUMENT NUMBER: 1988089133
TITLE: Analysis and genetic manipulation of Shigella virulence determinants for vaccine development.
AUTHOR: Mills S.D.; Sekizaki T.; Gonzalez-Carrero M.I.; Timmis K.N.
CORPORATE SOURCE: Department of Medical Biochemistry, University of Geneva, Geneva, Switzerland
SOURCE: Vaccine, (1988) 6/2 (116-122). ISSN: 0264-410X CODEN: VACCDE
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Shigellosis is a major public health problem in developing countries. Current epidemics of Shigella dysenteriae serotype 1 strains are particularly serious and are characterized by high mortality rates. A high proportion of the isolates are **resistant** to many of the antibiotics currently in use in these countries, a feature which seriously compromises clinical treatment of the infections. Efficacious vaccines are thus urgently needed. Basic studies on Shigella virulence factors, infections in laboratory models, and host responses has led to the development of several strategies for the production of vaccines. All of these are live oral vaccines involving bacteria capable of at least limited survival in the animal intestine and of carrying selected antigens to the mucosal immune system. One type of vaccine involves non-pathogenic shigellae, attenuated either by introduction of a requirement for aromatic

amino acids (aroD) or by loss of the large plasmid that specifies bacterial invasion of the mucosal epithelium. S. dysenteriae 1 strains under development as vaccines need to be engineered to eliminate high level **Shiga toxin** production, and a rapid and effective method to achieve this was recently elaborated. The second type of vaccine is represented by hybrid strains consisting of a carrier organism, such as an attenuated Salmonella or an Escherichia coli K-12 strain carrying the Shigella invasion plasmid, and the selected foreign antigen that it produces, in all cases so far the Shigella O antigen polysaccharide.

L12 ANSWER 121 OF 125 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 87203829 EMBASE
DOCUMENT NUMBER: 1987203829
TITLE: Production of a unique cytotoxin by Campylobacter jejuni.
AUTHOR: Guerrant R.L.; Wanke C.A.; Pennie R.A.; et al.
CORPORATE SOURCE: Division of Geographic Medicine, Department of Medicine, University of Virginia School of Medicine, Charlottesville, VA 22908, United States
SOURCE: Infection and Immunity, (1987) 55/10 (2526-2530). CODEN: INFIBR
COUNTRY: United States
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
052 Toxicology
LANGUAGE: English

AB Campylobacter jejuni is an important diarrheal pathogen worldwide; the mechanisms by which it causes disease remain unclear. Because of its association with inflammatory diarrhea, we postulated that C. jejuni might produce a cytotoxin similar to that produced by Shigella sp., enterohemorrhagic Escherichia coli O157, or Clostridium difficile. Filtrates of 12 polymyxin-treated isolates of C. jejuni were placed on HeLa cells (sensitive to **Shiga toxin** cytotoxicity) and Chinese hamster ovary (CHO) cells. Of 12 isolates of C. jejuni tested, 5 killed 50% of the cells at ≥1:4 dilutions of filtered suspensions of 109 bacteria per ml; killing was similar in HeLa and CHO cells (the CHO cells being **insensitive** to Shiga cytotoxin). One isolate produced a titer of 1:32 to 1:128. The relative potency in HeLa cells was comparable to that of E. coli strains that produce intermediate amounts of **Shiga**-like **toxin**. The other seven strains showed no cytotoxic effect, nor did the control diluents, polymyxin B, or supernatants of C. jejuni not treated with polymyxin B. Sonication also released active cytotoxin, but slightly less well than did polymyxin. The cytotoxic effect was dose dependent. Concentration of the C. jejuni in suspension by 10-fold before treatment with polymyxin B resulted in a 10-fold increase in the 50% cytotoxic dose. The cytotoxin effect was not neutralized by **Shiga toxin** immune serum against either **Shiga**-like **toxin** I or II or by anti-Clostridium difficile antiserum. The C. jejuni cytotoxin was partially labile to trypsin (0.25%) and to heating to ≥60°C. Cytotoxicity was retained in Scientific Products dialysis tubing D1615-1 (M(r) cutoff, 12,000 to 14,000). Some isolates of C. jejuni release a substance lethal to HeLa or CHO cells in vitro that is distinct from **Shiga**-like or Clostridium difficile **toxin**. This cytotoxin may contribute to the colonic mucosal invasive process that characterizes C. jejuni enteritis.

L12 ANSWER 122 OF 125 MEDLINE on STN DUPLICATE 33

ACCESSION NUMBER: 87306873 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3040592

TITLE: Localization of stx, a determinant essential for high-level production of shiga toxin by Shigella dysenteriae serotype 1, near pyrF and generation of stx transposon mutants.
AUTHOR: Sekizaki T; Harayama S; Brazil G M; Timmis K N
SOURCE: Infection and immunity, (1987 Sep) 55 (9) 2208-14.
Journal code: 0246127. ISSN: 0019-9567.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198709
ENTRY DATE: Entered STN: 19900305
Last Updated on STN: 19990129
Entered Medline: 19870930

AB Hfr strains of Shigella dysenteriae serotype 1 were constructed by transient integration of an RP4 plasmid derivative carrying transposon Tn501 into the Shigella chromosome through Tn501-mediated cointegration. The Hfr strains were mated with Escherichia coli K-12 recipients carrying various auxotrophic markers, and E. coli recombinants which had received prototrophic Shigella genes were selected. Some of the E. coli transconjugants produced high levels of a cytotoxin which was neutralized by both polyclonal and monoclonal anti-Shiga toxin sera. The determinant for Shiga toxin production, designated stx, was first transferred to E. coli K-12 and then mapped by Hfr crosses to the trp-pyrF region located at 30 min on the E. coli chromosome. Bacteriophage P1-mediated transduction analysis of stx gave the following gene order: trp-pyrF-stx. The level of Shiga toxin production in E. coli Stx+ transconjugants and transductants was as high as that of the parental S. dysenteriae 1 strain. Stx- mutants of an Stx+ E. coli transductant were generated by random in vivo insertion mutagenesis with a Tn10 derivative transposon, Tn-mini-kan, followed by P1 cotransduction of the kanamycin resistance and PyrF+ markers into a pyrF Stx+ E. coli K-12 recipient. One stx::Tn-mini-kan transposon mutation was transferred by P1 transduction from this E. coli Stx- mutant to an E. coli K-12 Hfr strain and in turn transferred by conjugation to the original S. dysenteriae 1 strain plus two others. All kanamycin-**resistant** recombinants of S. dysenteriae 1 had lost their ability to produce high levels of **Shiga toxin**. A gene that specifies high-level Shiga toxin production is thus located near pyrF on the chromosome of S. dysenteriae 1. Stx- mutants of S. dysenteriae 1 exhibited full virulence in the Sereny test.

L12 ANSWER 123 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1940:48651 CAPLUS
DOCUMENT NUMBER: 34:48651
ORIGINAL REFERENCE NO.: 34:7444d-f
TITLE: The relation between dysentery and the autonomic nervous system. I. Research on rabbits
AUTHOR(S): Kuroda, Hideo
SOURCE: Nagoya Igakkai Zasshi (1939), 49, 265 et seq.
CODEN: NYIZAK; ISSN: 0369-6723
DOCUMENT TYPE: Journal
LANGUAGE: Unavailable

AB Repeated injections of drugs together with severed vagus nerve (I) or severed sympathetic nerves (II) altered the **resistance** of the rabbits to **Shiga**-bacillus **toxin** (III) and to the Ohara bacillus (IV) as follows: atropine sulfate and I increased **resistance** especially against IV. Adrenaline chloride and I generally increased resistance but not invariably so for IV. Pilocarpine-HCl and acetylcholine and II decreased resistance. Adrenaline and I gave leucocytosis due to increased pseudoeosinophiles; there were no distinct changes in the other leucocytes or in the erythrocytes. With pilocarpine and II, the vagotonic blood picture, as found in man, did not

occur. Atropine and acetylcholine with I or II did not affect the blood picture.

L12 ANSWER 124 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1939:35568 CAPLUS
DOCUMENT NUMBER: 33:35568
ORIGINAL REFERENCE NO.: 33:5044f-g
TITLE: An experimental study on the relation between dysentery and vitamin C. II. The influence of vitamin C on the infection of rats with Ohara (Kruse-Sonne) bacilli; and the **resistance** of rabbits against **Shiga** [bacillus] **toxin** mixed with vitamin C
AUTHOR(S): Takahasi, Zyunzi
SOURCE: Nagoya Journal of Medical Science (1938), 12, Abstracts 51-2
CODEN: NJMSAG; ISSN: 0027-7622
DOCUMENT TYPE: Journal
LANGUAGE: German
AB In young rats injected with the Ohara dysentery bacillus by rectum, vitamin C given parenterally increased the resistance only slightly. Rabbits showed increased **resistance** to **Shiga** **toxin** mixed with vitamin C before administering.

L12 ANSWER 125 OF 125 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1939:35567 CAPLUS
DOCUMENT NUMBER: 33:35567
ORIGINAL REFERENCE NO.: 33:5044f-g
TITLE: An experimental study on the relation between dysentery and vitamin C. II. The influence of vitamin C on the infection of rats with Ohara (Kruse-Sonne) bacilli; and the **resistance** of rabbits against **Shiga** [bacillus] **toxin** mixed with vitamin C
AUTHOR(S): Takahasi, Zyunzi
SOURCE: Nagoya Igakkai Zasshi (1937), 46, 663
CODEN: NYIZAK; ISSN: 0369-6723
DOCUMENT TYPE: Journal
LANGUAGE: Unavailable
AB In young rats injected with the Ohara dysentery bacillus by rectum, vitamin C given parenterally increased the resistance only slightly. Rabbits showed increased **resistance** to **Shiga** **toxin** mixed with vitamin C before administering.

=> FIL STNGUIDE

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	140.81	223.90

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CA SUBSCRIBER PRICE	-3.50	-3.50

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=>
=> file medline biosis caplus embase wpids
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                         ENTRY    SESSION
FULL ESTIMATED COST                                       1.56     225.46

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)          SINCE FILE      TOTAL
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CA SUBSCRIBER PRICE                                       0.00      -3.50

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=> (mutated or mutant) and ((virulent or virulence or toxicity or toxic or
cytotoxic or cytotoxicity) 3N (increas? or more or great?))
MISSING OPERATOR TOXICITY) 3N
The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> (mutated or mutant) and ((virulent or virulence or toxicity or toxic or
cytotoxic or cytotoxicity) (3N) (increas? or more or great?))
   4 FILES SEARCHED...
L13       2725 (MUTATED OR MUTANT) AND ((VIRULENT OR VIRULENCE OR TOXICITY OR
               TOXIC OR CYTOTOXIC OR CYTOTOXICITY) (3N) (INCREAS? OR MORE OR
               GREAT?))

=> l13 and Shiga (3N) toxin
L14         15 L13 AND SHIGA (3N) TOXIN

=> dup rem l14
PROCESSING COMPLETED FOR L14
L15          6 DUP REM L14 (9 DUPLICATES REMOVED)

=> t ti l15 1-6

L15  ANSWER 1 OF 6     MEDLINE on STN                    DUPLICATE 1
TI   Activation of Shiga toxin type 2d (Stx2d) by elastase
     involves cleavage of the C-terminal two amino acids of the A2 peptide in
     the context of the appropriate B pentamer.

L15  ANSWER 2 OF 6  BIOSIS  COPYRIGHT (c) 2004 The Thomson Corporation.  on STN
TI   Association of the activatable Shiga toxin type 2
     variant, Stx2d, with an inducible bacteriophage.

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     on STN
TI   Comparison of the influence of mitomycin C on the in vivo virulaence of
     Shiga toxin producing Escherichia coli C600 (933W)
     lysogen and its isogenic recA mutants with EHEC O157:H7 strain 933 and its
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recA mutants in white mice.

L15 ANSWER 4 OF 6 MEDLINE on STN DUPLICATE 2
TI Role of the disulfide bond in **Shiga toxin** A-chain for toxin entry into cells.

L15 ANSWER 5 OF 6 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 3
TI Influence of mitomycin C on the virulence of Escherichia coli O157:H7 enterohemorrhagic strain 933 and its Rec A negative **mutant** strain in peroral infections of white mice

L15 ANSWER 6 OF 6 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 4
TI Use of the Vibrio cholerae irgA gene as a locus for insertion and expression of heterologous antigens in cholera vaccine strains.

=> d ibib abs l15 1-6

L15 ANSWER 1 OF 6 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2002124703 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11849548
TITLE: Activation of **Shiga toxin** type 2d (Stx2d) by elastase involves cleavage of the C-terminal two amino acids of the A2 peptide in the context of the appropriate B pentamer.
AUTHOR: Melton-Celsa Angela R; Kokai-Kun John F; O'Brien Alison D
CORPORATE SOURCE: Department of Microbiology, Uniformed Services University of the Health Sciences, F. Edward Hebert School of Medicine, 4301 Jones Bridge Road, Bethesda, MD 20814-4799, USA.
CONTRACT NUMBER: AI20148-17 (NIAID)
SOURCE: Molecular microbiology, (2002 Jan) 43 (1) 207-15. Journal code: 8712028. ISSN: 0950-382X.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200205
ENTRY DATE: Entered STN: 20020226
Last Updated on STN: 20020511
Entered Medline: 20020510

AB Shiga toxins (Stx) are potent ribosome-inactivating toxins that are produced by Shigella dysenteriae type 1 or certain strains of Escherichia coli. These toxins are composed of one A subunit that can be nicked and reduced to an enzymatically active A1(approximately 27 kDa) and an A2 peptide (approximately 4 kDa) as well as a pentamer of B subunits (approximately 7 kDa/monomer) that binds the eukaryotic cell. Purified **Shiga toxin** type 2d is activated 10- to 1000-fold for Vero cell toxicity by preincubation with mouse or human intestinal mucus or purified mouse elastase, whereas Stx2, Stx2c, Stx2e and Stx1 are not activatable. E. coli strains that produce the activatable Stx2d are **more virulent** in a streptomycin (str)-treated mouse model of infection [lethal dose 50% (LD50) = 101] than are E. coli strains that produce any other type of Stx (LD50 = 1010). To identify the element(s) of Stx2d that are required for mucus-mediated activation, toxin genes were constructed such that the expressed **mutant** toxins consisted of hybrids of Stx2d and Stx1, Stx2 or Stx2e, contained deletions of up to six amino acids from the C-terminus of the A2 of Stx2d or were altered in one or both of the two amino acids of the A2 of Stx2d that represent the only amino acid differences between the activatable Stx2d and the non-activatable Stx2c. Analysis of these **mutant** toxins

revealed that the A2 portion of Stx2d is required for toxin activation and that activation is abrogated if the Stx1 or Stx2e B subunit is substituted for the Stx2d B polypeptide. Furthermore, mass spectrometry performed on buffer- or elastase-treated Stx2d indicated that the A2 peptide of the activated Stx2d was two amino acids smaller than the A2 peptide from buffer-treated Stx2d. This finding, together with the toxin hybrid results, suggests that activation involves B pentamer-dependent cleavage by elastase of the C-terminal two amino acids from the Stx2d A2 peptide.

L15 ANSWER 2 OF 6 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2002:176497 BIOSIS
DOCUMENT NUMBER: PREV200200176497
TITLE: Association of the activatable **Shiga toxin** type 2 variant, Stx2d, with an inducible bacteriophage.
AUTHOR(S): Teel, L. D. [Reprint author]; Schmitt, C. K. [Reprint author]; Melton-Celsa, A. R. [Reprint author]; O'Brien, A. D. [Reprint author]
CORPORATE SOURCE: Uniformed Services University of the Health Sciences, Bethesda, MD, USA
SOURCE: Abstracts of the General Meeting of the American Society for Microbiology, (2001) Vol. 101, pp. 90-91. print. Meeting Info.: 101st General Meeting of the American Society for Microbiology. Orlando, FL, USA. May 20-24, 2001. American Society for Microbiology. ISSN: 1060-2011.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 6 Mar 2002
Last Updated on STN: 6 Mar 2002

AB The gene for **Shiga toxin** type 2 (Stx2) in Escherichia coli O157:H7 strains is typically borne on a readily inducible, toxin-converting lambdoid-like bacteriophage. However, the genes for variants of Stx2 present in **Shiga toxin**-producing Escherichia coli (STEC), with few exceptions, have not been demonstrated to be phage-borne. In this study, we examined the O91:H21 STEC isolate B2F1 that encodes two Stx2 variant toxins that are activatable by intestinal mucus for the presence of Stx2d-converting bacteriophages. First, we analyzed the DNA sequence of cosmids that encoded stx2d1 or stx2d2 and found that both toxin genes were flanked by similar sequences that resembled those published for Stx2 toxin-converting phages. Next, mutants of B2F1 were constructed that produced one or the other Stx2d toxin. Each **mutant** was then treated with an agent known to induce bacteriophages (mitomycin C). Toxin gene copy number increased in both mutants after mitomycin C treatment. However, toxin levels (as determined by Vero cell **cytotoxicity**) were substantially **increased** in sonically-disrupted extracts of the Stx2d1-producing **mutant** but not the Stx2d2-producing **mutant**. Furthermore, small turbid plaques were visible on a lawn of E. coli K-12 strain DH5a after induction of the Stx2d1-producing **mutant** and a putative stx2d1-containing lysogen was isolated. Induction of this lysogen that had been transformed with a RecA-expressing plasmid resulted in plaque formation and enhanced toxin production, indicators of the presence of an Stx2d1-converting phage. Finally, the pathogencity of the Stx2d1-producing **mutant** in the orally infected streptomycin-treated mouse model increased when animals were given subinhibitory doses of ciprofloxacin, a result which suggests increased toxin production by induction of the Stx2d1-converting bacteriophage lytic cycle in vivo. We conclude that only stx2d1 appears to be borne on an inducible toxin-converting bacteriophage and that stx2d2 expression is not influenced by bacteriophage induction.

L15 ANSWER 3 OF 6 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 1999243227 EMBASE

TITLE: Comparison of the influence of mitomycin C on the in vivo virulaence of **Shiga toxin** producing Escherichia coli C600 (933W) lysogen and its isogenic recA mutants with EHEC O157:H7 strain 933 and its recA mutants in white mice.

AUTHOR: Alexiev R.; Muhldorfer I.; Nenkov P.; Hacker J.

CORPORATE SOURCE: R. Alexiev, NCIPD, 26 Yanko Sakazov Blvd., 1504 Sofia, Bulgaria. twins@main.infotel.bg

SOURCE: Problems of Infectious and Parasitic Diseases, (1998) 26/SUPPL. (5-7).
Refs: 20
ISSN: 0204-9155 CODEN: PIPDD4

COUNTRY: Bulgaria

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology
005 General Pathology and Pathological Anatomy
016 Cancer
028 Urology and Nephrology
037 Drug Literature Index

LANGUAGE: English

SUMMARY LANGUAGE: English

AB **Shiga toxin** producing enterohemorrhagic E. coil (EHEC) strains cause diarrhoea, hemorrhagic colitis and the hemolytic uremic syndrome. Three week old ICR mice were treated perorally with streptomycin and intraperitoneally with mitomycin C, and consecutively infected intragastrally with either of the recA+ or isogenic recA mutants of the enterohemorrhagic E. coli 0157:H7 strain 933, carrying the Stx1 and Stx2 converting phages 933J and 933W, respecively, or the E coli K-12 derivative C600(933W), carrying only the Stxa converting phage. Following infection with the recA+ strains weight loss was observed as well as bending of the spinal column and partial or total paralysis with consequent death. The administration of mitomycin C resulted in an **increased virulence** of the recA+ EHEC strain 933 and E. coli strain C600(933W) by 80% and 60%, respectively, compared to the control group of mice which were infected with the same strain hut were not subjected to treatment with this antibiotic. The animals infected with the recA mutants and treated with different doses of mitomycin C showed no symptoms of disease and no mortality in all groups of mice. The recA mutants did not induce any pathological changes or death in the living organisms. The experiments confirmed the role of **Shiga toxin** for the in vivo virulence of the recA+ E. coil strains in animals and explain a role of Stx in the development of cancer associated hemolytic uremic syndrome (cHUS) following clinical treatment of patients suffering neoplasia with mitomycin C.

L15 ANSWER 4 OF 6 MEDLINE on STN DUPLICATE 2

ACCESSION NUMBER: 97269051 MEDLINE

DOCUMENT NUMBER: PubMed ID: 9111051

TITLE: Role of the disulfide bond in **Shiga toxin** A-chain for toxin entry into cells.

AUTHOR: Garred O; Dubinina E; Polesskaya A; Olsnes S; Kozlov J; Sandvig K

CORPORATE SOURCE: Institute for Cancer Research at The Norwegian Radium Hospital, Montebello, 0310 Oslo, Norway.

SOURCE: Journal of biological chemistry, (1997 Apr 25) 272 (17) 11414-9.
Journal code: 2985121R. ISSN: 0021-9258.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199705
ENTRY DATE: Entered STN: 19970602
Last Updated on STN: 20000303
Entered Medline: 19970521

AB **Shiga toxin** consists of an enzymatically active A-chain and a pentameric binding subunit. The A-chain has a trypsin-sensitive region, and upon cleavage two disulfide bonded fragments, A1 and A2, are generated. To study the role of the disulfide bond, it was eliminated by mutating cysteine 242 to serine. In T47D cells this **mutated** toxin was **more toxic** than wild type toxin after a short incubation, whereas after longer incubation times wild type toxin was most toxic. Cells cleaved not only wild type but also **mutated** A-chain into A1 and A2 fragments. The **mutated** A-chain was more sensitive than wild type toxin to Pronase, and it was degraded at a higher rate in T47D cells. Subcellular fractionation demonstrated transport of both wild type and **mutated** toxin to the Golgi apparatus. Brefeldin A, which disrupts the Golgi apparatus, protected not only against **Shiga toxin** but also against the **mutated** toxin, indicating involvement of the Golgi apparatus. After prebinding of **Shiga**(C242S) **toxin** to wells coated with the **Shiga toxin** receptor, Gb3, trypsin treatment induced dissociation of A1 from the toxin-receptor complex demonstrating that in addition to stabilizing the A-chain, the disulfide bond prevents dissociation of the A1 fragment from the toxin-receptor complex.

L15 ANSWER 5 OF 6 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 3
ACCESSION NUMBER: 1998:212371 CAPLUS
DOCUMENT NUMBER: 128:289781
TITLE: Influence of mitomycin C on the virulence of Escherichia coli O157:H7 enterohemorrhagic strain 933 and its Rec A negative **mutant** strain in peroral infections of white mice
AUTHOR(S): Aleksiev, R.; Nenkov, P.; Muhldorfer, I.; Hacker, I.
CORPORATE SOURCE: National Institute of Infectious and Parasitic Diseases, Sofia, 1504, Bulg.
SOURCE: Problems of Infectious and Parasitic Diseases (1997), 24(2), 32-36
CODEN: PIPDD4; ISSN: 0204-9155
PUBLISHER: National Center of Infectious and Parasitic Diseases
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The enterohemorrhagic strains of Escherichia coli cause diarrhea, hemorrhagic colitis and hemorrhagic uremic syndrome. Most of them synthesize **Shiga**-like **toxin** II. Three week old ICR mice treated perorally with streptomycin and interperitoneally with Mitomycin C were infected intragastrically with an E. coli O157:H7 enterohemorrhagic strain 933 and its Rec A neg. **mutant** stain producing insignificant quantities of toxin. After infection with Rec apos. stain 933 weight loss was observed as well as visually exhibited bending of the spinal column and partial of total paralysis with consequent death. The administered Micomycin C **increased virulence** of the Rec A pos. strain by 80% compared to the control group of mice which were infected with the same strain but were not subjected to treatment with antibiotics. The animals infected with different doses genetically engineered Rec a neg. strain 933 and treated with different doses Mitomycin C showed no symptoms of disease and no mortality in all groups of mice. After destruction of the Rec a gene in the initial strain the acquired **mutant** strain was practically avirulent and did not

induce pathol. changes or death in the living organisms. The performed expts. partially unveiled the mechanisms of regulation of toxin production in enterohemorrhagic E. coli strains.

REFERENCE COUNT: 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 6 OF 6 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
DUPLICATE 4

ACCESSION NUMBER: 1994:18123 BIOSIS
DOCUMENT NUMBER: PREV199497031123
TITLE: Use of the Vibrio cholerae irgA gene as a locus for insertion and expression of heterologous antigens in cholera vaccine strains.
AUTHOR(S): Butterton, Joan R.; Boyko, Stephanie A.; Calderwood, Stephen B. [Reprint author]
CORPORATE SOURCE: Dep. Microbiol. Mol. Genet., Harvard Med. Sch., Boston, MA 02115, USA
SOURCE: Vaccine, (1993) Vol. 11, No. 13, pp. 1327-1335. CODEN: VACCDE. ISSN: 0264-410X.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 25 Jan 1994
Last Updated on STN: 25 Jan 1994

AB Vibrio cholerae may be a particularly effective organism for use in delivering heterologous antigens to stimulate a common mucosal immune response. A live attenuated vaccine strain of V. cholerae was constructed from the ctxA deletion **mutant** 0395-N1, containing the B subunit of **Shiga**-like **toxin** I under the transcriptional control of the iron-regulated irgA promoter. The B subunit of **Shiga**-like **toxin** I is identical to the B subunit of **Shiga toxin** (StxB). irgA encodes the major iron-regulated outer membrane protein of V. cholerae, which is a known virulence factor for this organism. Clones of the structural gene irgA from the classical V. cholerae strain 0395, with the gene for the **Shiga**-like **toxin** I B subunit inserted under the control of the irgA promoter, were used to introduce an internal deletion of irgA into the chromosome of 0395-N1 by in vivo marker exchange, using the suicide vector plasmid pCVD442. This plasmid contains the sacB gene from Bacillus subtilis, which allowed positive selection for loss of plasmid sequences on exposure to sucrose. The construction of vaccine strains was confirmed by Southern hybridization studies and outer membrane protein analysis. The expression of StxB in the vaccine strain VAC2 following growth in high- or low-iron conditions was shown to be tightly iron-regulated by Western blot analysis and by quantification of StxB using a sandwich enzyme-linked immunosorbent assay. The production of StxB by VAC2 under low-iron conditions was greater than that of the reference strain Shigella dysenteriae 60R. This vaccine strain produced no detectable cytotoxicity in a HeLa cell assay, and showed no **increased virulence** over the attenuated parent strain, 0395-N1, in a suckling mouse model. We suggest that the V. cholerae irgA gene is a particularly useful locus for the insertion and expression of heterologous antigens in cholera vaccine strains for oral delivery.

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FULL ESTIMATED COST	42.88	268.34

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     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1             0 SHIGA 5A TOXIN
L2           193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3             1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4         20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5           182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6           122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7            16 L2 AND L6
L8            10 DUP REM L7 (6 DUPLICATES REMOVED)

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     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18
     NOV 2004
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L10            2 DUP REM L9 (0 DUPLICATES REMOVED)
L11            1 L2 AND L4
L12          125 DUP REM L2 (68 DUPLICATES REMOVED)

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     NOV 2004
L13         2725 (MUTATED OR MUTANT) AND ((VIRULENT OR VIRULENCE OR TOXICITY OR
L14           15 L13 AND SHIGA (3N) TOXIN
L15            6 DUP REM L14 (9 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 17:03:25 ON 18 NOV 2004

=> (ricin or abrin or gelonin or crotin or pokeweed (w) antiviral (w) protein or
saporin or momordin or modeccin or sarcin or diphtheria (w) toxin or exotoxin (w) A
UNMATCHED LEFT PARENTHESIS '(RICIN'
The number of right parentheses in a query must be equal to the
number of left parentheses.

=> ricin or abrin or gelonin or crotin or pokeweed (w) antiviral (w) protein or
saporin or momordin or modeccin or sarcin or diphtheria (w) toxin or exotoxin (w) A
             0 RICIN
             0 ABRIN
             0 GELONIN
             0 CROTIN
             0 POKEWEED
             0 ANTIVIRAL
             7 PROTEIN
             0 POKEWEED (W) ANTIVIRAL (W) PROTEIN
             0 SAPORIN
             0 MOMORDIN
             0 MODECCIN
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              0 SARCIN
              0 DIPHTHERIA
              0 TOXIN
              0 DIPHTHERIA (W) TOXIN
              0 EXOTOXIN
            234 A
              0 EXOTOXIN (W) A
L16           0 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL
                (W) PROTEIN OR SAPORIN OR MOMORDIN OR MODECCIN OR SARCIN OR
                DIPHTHERIA (W) TOXIN OR EXOTOXIN (W) A

=> file medline biosis caplus embase wpids
COST IN U.S. DOLLARS                                  SINCE FILE      TOTAL
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FULL ESTIMATED COST                                       1.68     270.02

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                                                         ENTRY    SESSION
CA SUBSCRIBER PRICE                                       0.00      -4.20

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=> ricin or abrin or gelonin or crotin or pokeweed (w) antiviral (w) protein or
saporin or momordin or modeccin or sarcin or diphtheria (w) toxin or exotoxin (w) A
L17       32942 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL
                (W) PROTEIN OR SAPORIN OR MOMORDIN OR MODECCIN OR SARCIN OR
                DIPHTHERIA (W) TOXIN OR EXOTOXIN (W) A

=> d his

     (FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1            0 SHIGA 5A TOXIN
L2          193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3            1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4        20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5          182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6          122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7           16 L2 AND L6
L8           10 DUP REM L7 (6 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18
     NOV 2004
L9            2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
```

L10 2 DUP REM L9 (0 DUPLICATES REMOVED)
L11 1 L2 AND L4
L12 125 DUP REM L2 (68 DUPLICATES REMOVED)

FILE 'STNGUIDE' ENTERED AT 16:42:37 ON 18 NOV 2004

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:58:30 ON 18 NOV 2004
L13 2725 (MUTATED OR MUTANT) AND ((VIRULENT OR VIRULENCE OR TOXICITY OR
L14 15 L13 AND SHIGA (3N) TOXIN
L15 6 DUP REM L14 (9 DUPLICATES REMOVED)

FILE 'STNGUIDE' ENTERED AT 17:03:25 ON 18 NOV 2004
L16 0 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 17:19:55 ON 18 NOV 2004
L17 32942 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (

=> l13 and l17
L18 108 L13 AND L17

=> dup rem l18
PROCESSING COMPLETED FOR L18
L19 41 DUP REM L18 (67 DUPLICATES REMOVED)

=> t ti l19 1-41

L19 ANSWER 1 OF 41 MEDLINE on STN DUPLICATE 1
TI Expression of **ricin** A chain and **ricin** A chain-KDEL in Escherichia coli.

L19 ANSWER 2 OF 41 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI **Mutant** bispecific antibody which is rapidly cleared from a patient's body, useful as a pretargeting agent in methods for diagnosing or treating diseases, such as autoimmune disease, and disease caused by infectious agents.

L19 ANSWER 3 OF 41 MEDLINE on STN DUPLICATE 2
TI Structural analysis by X-ray crystallography and calorimetry of a haemagglutinin component (HA1) of the progenitor toxin from Clostridium botulinum.

L19 ANSWER 4 OF 41 MEDLINE on STN DUPLICATE 3
TI Extracellular Bad fused to toxin transport domains induces apoptosis.

L19 ANSWER 5 OF 41 MEDLINE on STN DUPLICATE 4
TI Targeted therapy against human lung cancer in nude mice by high-affinity recombinant antimesothelin single-chain Fv immunotoxin.

L19 ANSWER 6 OF 41 MEDLINE on STN DUPLICATE 5
TI [Endosomes and toxin translocation].
Endosomes et translocation de toxines.

L19 ANSWER 7 OF 41 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Treating disease with immunotoxin, useful against leukemia, lymphoma and myeloma, after treatment to upregulate the target antigen.

L19 ANSWER 8 OF 41 WPIDS COPYRIGHT 2004 THE THOMSON CORP on STN
TI Inhibiting rejection response in a primate recipient to foreign cells involves exposing the recipient to anti-CD3-DT immunotoxin with reduced anti-DT antibody binding and then transplanting the donor cells.

L19 ANSWER 9 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Immunotoxins with increased activity against epidermal growth factor receptor vIII-expressing cells produced by antibody phage display

L19 ANSWER 10 OF 41 MEDLINE on STN DUPLICATE 6
TI Expression of an anti-CD3 single-chain immunotoxin with a truncated **diphtheria toxin** in a **mutant** CHO cell line.

L19 ANSWER 11 OF 41 MEDLINE on STN DUPLICATE 7
TI Dependence of **ricin** toxicity on translocation of the toxin A-chain from the endoplasmic reticulum to the cytosol.

L19 ANSWER 12 OF 41 MEDLINE on STN DUPLICATE 8
TI A deletion within the translocation domain of Pseudomonas **exotoxin A** enhances translocation efficiency and cytotoxicity concomitantly.

L19 ANSWER 13 OF 41 MEDLINE on STN DUPLICATE 9
TI Amino acid substitution in alpha-helix 7 of Cry1Ac delta-endotoxin of Bacillus thuringiensis leads to enhanced toxicity to Helicoverpa armigera Hubner.

L19 ANSWER 14 OF 41 MEDLINE on STN DUPLICATE 10
TI Modification of **ricin** A chain, by addition of endoplasmic reticulum (KDEL) or Golgi (YQRL) retention sequences, enhances its cytotoxicity and translocation.

L19 ANSWER 15 OF 41 MEDLINE on STN
TI Furin-mediated cleavage of Pseudomonas exotoxin-derived chimeric toxins.

L19 ANSWER 16 OF 41 MEDLINE on STN DUPLICATE 11
TI **Ricin** toxin contains at least three galactose-binding sites located in B chain subdomains 1 alpha, 1 beta, and 2 gamma.

L19 ANSWER 17 OF 41 MEDLINE on STN DUPLICATE 12
TI The reaction of bacterial toxins with formaldehyde and its use for antigen stabilization.

L19 ANSWER 18 OF 41 MEDLINE on STN DUPLICATE 13
TI Lec32 is a new mutation in Chinese hamster ovary cells that essentially abrogates CMP-N-acetylneuraminic acid synthetase activity.

L19 ANSWER 19 OF 41 MEDLINE on STN DUPLICATE 14
TI An anti-CD3 single-chain immunotoxin with a truncated **diphtheria toxin** avoids inhibition by pre-existing antibodies in human blood.

L19 ANSWER 20 OF 41 MEDLINE on STN DUPLICATE 15
TI Ceramide reverses brefeldin A (BFA) resistance in BFA-resistant cell lines.

L19 ANSWER 21 OF 41 MEDLINE on STN DUPLICATE 16
TI Role of **exotoxin A** in inducing severe Pseudomonas aeruginosa infections in mice.

L19 ANSWER 22 OF 41 MEDLINE on STN DUPLICATE 17
TI Cleavage of pseudomonas exotoxin and **diphtheria toxin** by a furin-like enzyme prepared from beef liver.

L19 ANSWER 23 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Site-Specific Conjugation to Interleukin 4 Containing **Mutated** Cysteine Residues Produces Interleukin 4-Toxin Conjugates with Improved

Binding and Activity

L19 ANSWER 24 OF 41 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 18
TI An immunotoxin with increased activity and homogeneity produced by reducing the number of lysine residues in recombinant Pseudomonas exotoxin.

L19 ANSWER 25 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI **Mutant** cytokines having increased receptor affinity

L19 ANSWER 26 OF 41 MEDLINE on STN DUPLICATE 19
TI Heparin-binding transforming growth factor alpha-Pseudomonas **exotoxin A**. A heparan sulfate-modulated recombinant toxin cytotoxic to cancer cells and proliferating smooth muscle cells.

L19 ANSWER 27 OF 41 MEDLINE on STN DUPLICATE 20
TI Alanine scanning mutagenesis identifies surface amino acids on domain II of Pseudomonas exotoxin required for cytotoxicity, proper folding, and secretion into periplasm.

L19 ANSWER 28 OF 41 MEDLINE on STN DUPLICATE 21
TI A recombinant form of Pseudomonas exotoxin directed at the epidermal growth factor receptor that is cytotoxic without requiring proteolytic processing.

L19 ANSWER 29 OF 41 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Interactions of **diphtheria toxin** B-fragment with cells. Role of amino- and carboxyl-terminal regions.

L19 ANSWER 30 OF 41 MEDLINE on STN DUPLICATE 22
TI Phe496 and Leu497 are essential for receptor binding and cytotoxic action of the murine interleukin-4 receptor targeted fusion toxin DAB389-mIL-4.

L19 ANSWER 31 OF 41 MEDLINE on STN DUPLICATE 23
TI A proper amino terminus of **diphtheria toxin** is important for cytotoxicity.

L19 ANSWER 32 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Enhancement of the cytotoxicity of mistletoe lectin-1 (ML-1) by high pH or perturbation in Golgi functions

L19 ANSWER 33 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Cytotoxic recombinant Pseudomonas endotoxin and target-specific fusion products

L19 ANSWER 34 OF 41 MEDLINE on STN DUPLICATE 24
TI **Increased cytotoxicity** of **ricin** in a putative Golgi-defective **mutant** of Chinese hamster ovary cell.

L19 ANSWER 35 OF 41 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 25
TI DOMAIN II MUTANTS OF PSEUDOMONAS EXOTOXIN DEFICIENT IN TRANSLOCATION.

L19 ANSWER 36 OF 41 MEDLINE on STN
TI Restoration of enzymic activity and cytotoxicity of **mutant**, E553C, Pseudomonas aeruginosa **exotoxin A** by reaction with iodoacetic acid.

L19 ANSWER 37 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Antibody-DT conjugates in the study of surface antigen properties and DT

cytotoxic mechanism

L19 ANSWER 38 OF 41 MEDLINE on STN DUPLICATE 26
TI Properties of baby-hamster kidney (BHK) cells treated with Swainsonine, an inhibitor of glycoprotein processing. Comparison with **ricin** -resistant BHK-cell mutants.

L19 ANSWER 39 OF 41 MEDLINE on STN
TI **Increased cytotoxicity** of normal rabbit serum for lectin-resistant mutants of animal cells.

L19 ANSWER 40 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
TI Formation of a hybrid toxin from **ricin** agglutinin and a non-toxic **mutant** protein of **diphtheria toxin**

L19 ANSWER 41 OF 41 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
TI Ricinus communis toxin mediated inhibition of protein synthesis in cell free extracts of a toxin resistant variant mouse lymphoma cell line.

=> d ibib abs l19 9-16, 21, 24-36, 38, 40,41

L19 ANSWER 9 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2000:573473 CAPLUS
DOCUMENT NUMBER: 134:141422
TITLE: Immunotoxins with increased activity against epidermal growth factor receptor vIII-expressing cells produced by antibody phage display
AUTHOR(S): Beers, Richard; Chowdhury, Partha; Bigner, Darell; Pastan, Ira
CORPORATE SOURCE: Laboratory of Molecular Biology, National Cancer Institute, NIH, Bethesda, MD, 20892, USA
SOURCE: Clinical Cancer Research (2000), 6(7), 2835-2843
CODEN: CCREF4; ISSN: 1078-0432
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Recombinant immunotoxins are fusion proteins composed of Fv regions of antibodies and bacterial or plant toxins that are being developed for the targeted therapy of cancer. MR1(Fv)-PE38 is a single-chain recombinant immunotoxin that targets a **mutant** form of the epidermal growth factor receptor (EGFR), EGFRvIII, that is frequently over-expressed in malignant glioblastomas. The authors used random complementarity determining region (CDR) mutagenesis to obtain mutants of MR1(Fv) with an increased affinity for EGFRvIII and an increased activity when converted to a recombinant immunotoxin. Initially, 9 residues of heavy chain CDR3 were randomly mutagenized, and several mutants with increased binding affinity were isolated. All mutations were located at amino acids 98 and 99, which correspond to a DNA hot spot, a DNA sequence that mutates at high frequency during natural antibody maturation. A specific region of variable region of antibody light chain CDR3 was mutagenized that corresponded to a hot spot and a **mutant** (MR1-1) with an addnl. **increase** in affinity, and **cytotoxic** activity was isolated. Thus, targeting hot spots in the CDRs of Fvs is an effective approach to obtaining Fvs with increased affinity. The increased affinity of MR1-1(Fv) makes it an attractive candidate for the targeted therapy of glioblastomas.
REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 10 OF 41 MEDLINE on STN DUPLICATE 6

ACCESSION NUMBER: 2000334448 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10873546
TITLE: Expression of an anti-CD3 single-chain immunotoxin with a truncated **diphtheria toxin** in a **mutant** CHO cell line.
AUTHOR: Liu Y Y; Gordienko I; Mathias A; Ma S; Thompson J; Woo J H; Neville D M Jr
CORPORATE SOURCE: Section on Biophysical Chemistry, National Institute of Mental Health, Bethesda, Maryland, 28092-4034, USA.
SOURCE: Protein expression and purification, (2000 Jul) 19 (2) 304-11.
Journal code: 9101496. ISSN: 1046-5928.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200008
ENTRY DATE: Entered STN: 20000811
Last Updated on STN: 20000811
Entered Medline: 20000802

AB ADP-ribosylating immunotoxins are generally expressed in Escherichia coli and then refolded in vitro. Because the efficiency of the in vitro refolding process decreases with the number of protein domains and internal disulfide bonds, these immunotoxins have been generally limited to single-chain monovalent structures. We now show that using the hamster cell line CHO K1 RE1.22c (J. M. Moehring and T. J. Moehring, 1979, Somat. Cell Genet. 5, 453-468) that has been **mutated** to ADP-ribosylation insensitivity, a level of 4 microg/ml of a truncated anti-T cell immunotoxin, DT390-scFvUCHT1, can be secreted into the medium. This immunotoxin is glycosylated at the two potential N-linked glycosylation sites in the toxin moiety: positions 16-18 in the A chain and residues 235-237 in the B chain. The glycosylated immunotoxin is relatively nontoxic (IC(50) 4.8 x 10(-10) M). Removal of the N-linked oligosaccharides by N-glycosidase F treatment or mutations at the two N-linked glycosylation sites results in a highly active immunotoxin with an IC(50) of 4 x 10(-12) M toward CD3(+) Jurkat cells. This is a 12-fold **increase** in **toxicity** over the same immunotoxin harvested from E. coli periplasm without refolding. A single Asn(235) Ala mutation that removed the B chain glycosylation was nearly as toxic as the double **mutant**. This suggests that B chain glycosylation is the major cause for the loss of toxicity.

L19 ANSWER 11 OF 41 MEDLINE on STN DUPLICATE 7
ACCESSION NUMBER: 2000036595 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10567425
TITLE: Dependence of **ricin** toxicity on translocation of the toxin A-chain from the endoplasmic reticulum to the cytosol.
AUTHOR: Wesche J; Rapak A; Olsnes S
CORPORATE SOURCE: Institute for Cancer Research, The Norwegian Radium Hospital, Montebello, 0310 Oslo, Norway.
SOURCE: Journal of biological chemistry, (1999 Nov 26) 274 (48) 34443-9.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199912
ENTRY DATE: Entered STN: 20000113
Last Updated on STN: 20000113

Entered Medline: 19991229

AB **Ricin** acts by translocating to the cytosol the enzymatically active toxin A-chain, which inactivates ribosomes. Retrograde intracellular transport and translocation of **ricin** was studied under conditions that alter the sensitivity of cells to the toxin. For this purpose tyrosine sulfation of **mutant** A-chain in the Golgi apparatus, glycosylation in the endoplasmic reticulum (ER) and appearance of A-chain in the cytosolic fraction was monitored. Introduction of an ER retrieval signal, a C-terminal KDEL sequence, into the A-chain **increased** the **toxicity** and resulted in **more** efficient glycosylation, indicating enhanced transport from Golgi to ER. Calcium depletion inhibited neither sulfation nor glycosylation but inhibited translocation and toxicity, suggesting that the toxin is translocated to the cytosol by the pathway used by misfolded proteins that are targeted to the proteasomes for degradation. Slightly acidified medium had a similar effect. The proteasome inhibitor, lactacystin, sensitized cells to **ricin** and increased the amount of **ricin** A-chain in the cytosol. Anti-Sec61alpha precipitated sulfated and glycosylated **ricin** A-chain, suggesting that retrograde toxin translocation involves Sec61p. The data indicate that retrograde translocation across the ER membrane is required for intoxication.

L19 ANSWER 12 OF 41 MEDLINE on STN DUPLICATE 8

ACCESSION NUMBER: 1999217016 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10200959
TITLE: A deletion within the translocation domain of Pseudomonas **exotoxin A** enhances translocation efficiency and cytotoxicity concomitantly.
AUTHOR: Taupiac M P; Bebien M; Alami M; Beaumelle B
CORPORATE SOURCE: Departement Biologie-Sante, Universite Montpellier II, France.
SOURCE: Molecular microbiology, (1999 Mar) 31 (5) 1385-93. Journal code: 8712028. ISSN: 0950-382X.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199907
ENTRY DATE: Entered STN: 19990715
Last Updated on STN: 20020420
Entered Medline: 19990706

AB Pseudomonas **exotoxin A** (PE) is a cytotoxin composed of three structural domains. Domain I is responsible for cell binding, domain II for membrane translocation enabling access to the cytosol, and domain III for the catalytic inactivation of protein synthesis, which results in cell death. To investigate the role of the six alpha-helices (A-F) that form the translocation domain, we deleted them successively one at a time. All mutants showed native cell-binding and catalytic activities, indicating that deletions specifically affected translocation activity. This step of the intoxication procedure was examined directly using a cell-free translocation assay, and indirectly by monitoring cytotoxicity. Translocation activity and log(cytotoxicity) were highly correlated, directly indicating that translocation is rate limiting for PE intoxication. Deletion of B, C and D helices resulted in non-toxic and non-translocating molecules, whereas mutants lacking the A or E helix displayed significant cytotoxicity albeit 500-fold lower than native PE. We concluded that B, C and D helices, which make up the core of domain II, are essential, whereas the more peripheral A and E helices are comparatively dispensable. The last helix (F) is inhibitory for translocation because its deletion produced a **mutant** displaying a translocation activity 60% higher than PE, along with a three- to

sixfold **increase** in **cytotoxicity** in all tested cell lines. This toxin is the most in vitro active PE **mutant** obtained until now. Finally, partial duplication of domain II did not give rise to a more actively translocated PE, but rather to a threefold less active molecule.

L19 ANSWER 13 OF 41 MEDLINE on STN DUPLICATE 9
ACCESSION NUMBER: 1999412187 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10481060
TITLE: Amino acid substitution in alpha-helix 7 of Cry1Ac delta-endotoxin of Bacillus thuringiensis leads to enhanced toxicity to Helicoverpa armigera Hubner.
AUTHOR: Chandra A; Ghosh P; Mandaokar A D; Bera A K; Sharma R P; Das S; Kumar P A
CORPORATE SOURCE: National Research Centre for Plant Biotechnology, Indian Agricultural Research Institute, New Delhi.
SOURCE: FEBS letters, (1999 Sep 17) 458 (2) 175-9.
Journal code: 0155157. ISSN: 0014-5793.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199910
ENTRY DATE: Entered STN: 19991026
Last Updated on STN: 19991026
Entered Medline: 19991013

AB Insecticidal proteins or delta-endotoxins of Bacillus thuringiensis are highly toxic to a wide range of agronomically important pests. The toxins are formed of three structural domains. The N-terminal domain is a bundle of eight alpha-helices and is implicated in pore formation in insect midgut epithelial membranes. All the delta-endotoxins share a common hydrophobic motif of eight amino acids in alpha-helix 7. A similar motif is also present in fragment B of **diphtheria toxin** (DT). Site-directed mutagenesis of Cry1Ac delta-endotoxin of B. thuringiensis was carried out to substitute its hydrophobic motif with that of DT fragment B. The **mutant** toxin was shown to be **more toxic** to the larvae of Helicoverpa armigera (cotton bollworm) than the wild-type toxin. Voltage clamp analysis with planar lipid bilayers revealed that the **mutant** toxin opens larger ion channels and induces higher levels of conductance than the wild-type toxin.

L19 ANSWER 14 OF 41 MEDLINE on STN DUPLICATE 10
ACCESSION NUMBER: 1998179116 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9520293
TITLE: Modification of **ricin** A chain, by addition of endoplasmic reticulum (KDEL) or Golgi (YQRL) retention sequences, enhances its cytotoxicity and translocation.
AUTHOR: Zhan J; Stayton P; Press O W
CORPORATE SOURCE: Department of Medicine, University of Washington, Seattle 98195, USA.
CONTRACT NUMBER: R01 CA55596 (NCI)
SOURCE: Cancer immunology, immunotherapy : CII, (1998 Mar) 46 (1) 55-60.
Journal code: 8605732. ISSN: 0340-7004.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199804
ENTRY DATE: Entered STN: 19980416
Last Updated on STN: 19980416

Entered Medline: 19980408

AB A pKK expression system in Escherichia coli was used to produce recombinant **ricin** A chain (rRTA) and rRTA modified by addition of organelle-specific amino acid retention sequences, including KDEL (an endoplasmic reticulum, ER, lumen retention signal), KKMP (an ER membrane retention signal), YQRL (a trans-Golgi network retention signal) and KFERQ (a lysosome-targeting signal) to the C terminus of rRTA. The toxicities of these RTA mutants were assessed in Jurkat cells following fluid-phase endocytosis. rRTA-KDEL and rRTA-YQRL were significantly **more cytotoxic** for Jurkat cells than rRTA, rRTA-KKMP or rRTA-KFERQ. This difference did not result from signal(KDEL or YQRL)-mediated binding of these RTA mutants to the cell surface. Reconstituted ER and Golgi vesicles have been employed to assess translocation of rRTA and **mutant** rRTA. RTA-KDEL and RTA-YQRL respectively exhibited 6.7-fold and 6.1-fold more protection against papain digestion in reconstituted ER vesicles and 2.2-fold and 1.8-fold more protection in reconstituted Golgi vesicles, than unmodified rRTA. These mutants were reassociated with **ricin** B chain to form holotoxins. The **mutant** RTA-KDEL and RTA-YQRL holotoxins were 3.8-fold and 1.5-fold **more cytotoxic** for target cells, respectively, than **ricin** produced using unmodified rRTA. Our results suggest that both ER and the trans-Golgi network may play important roles in the intracellular trafficking and translocation of **ricin** A chain.

L19 ANSWER 15 OF 41 MEDLINE on STN

ACCESSION NUMBER: 1998058966 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9395513
TITLE: Furin-mediated cleavage of Pseudomonas exotoxin-derived chimeric toxins.
AUTHOR: Chiron M F; Fryling C M; FitzGerald D
CORPORATE SOURCE: Biotherapy Section, Laboratory of Molecular Biology, DBS, NCI, National Institutes of Health, Bethesda, Maryland 20892, USA.
SOURCE: Journal of biological chemistry, (1997 Dec 12) 272 (50) 31707-11.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199801
ENTRY DATE: Entered STN: 19980129
Last Updated on STN: 19980129
Entered Medline: 19980115

AB Pseudomonas exotoxin (PE) requires proteolytic cleavage to generate a 37-kDa C-terminal fragment that translocates to the cytosol and ADP-ribosylates elongation factor 2. Cleavage within cells is mediated by furin, occurs between arginine 279 and glycine 280, and requires an arginine at both P1 and P4 residues. To study the proteolytic processing of PE-derived chimeric toxins, TGFalpha-PE38 (transforming growth factor fused to the domains II and III of PE) and a **mutant** form, TGFalpha-PE38gly279, were each produced in Escherichia coli. When assessed on various epidermal growth factor (EGF) receptor-positive cell lines, TGFalpha-PE38 was 100-500-fold **more toxic** than TGFalpha-PE38gly279. In contrast to PE, where cleavage by furin is only evident at pH 5.5, furin cleaved TGFalpha-PE38 over a broad pH range, while TGFalpha-PE38gly279 was resistant to cleavage. TGFalpha-PE38 was poorly toxic for furin-deficient LoVo cells, unless it was first pretreated in vitro with furin. Furin treatment produced a nicked protein that was 30-fold **more toxic** than its unnicked counterpart. Using the single chain immunotoxin HB21scFv-PE40 as a substrate, furin-mediated processing of an antibody-based immunotoxin was

also evaluated. HB21scFv-PE40, which targets cells expressing the transferrin receptor, was cleaved in a similar fashion to that of TGFalpha-PE38 and nicked HB21scFv-PE40 exhibited **increased toxicity** for LoVo cells. In short-term experiments, the rate of reduction in protein synthesis by furin-nicked immunotoxins was increased compared with unnicked protein, indicating that cleavage by furin can be a rate-limiting step. We conclude that furin-mediated cleavage of PE-derived immunotoxins is important for their cytotoxic activity.

L19 ANSWER 16 OF 41 MEDLINE on STN DUPLICATE 11
ACCESSION NUMBER: 97098095 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8942636
TITLE: **Ricin** toxin contains at least three galactose-binding sites located in B chain subdomains 1 alpha, 1 beta, and 2 gamma.
AUTHOR: Frankel A E; Burbage C; Fu T; Tagge E; Chandler J; Willingham M C
CORPORATE SOURCE: Department of Medicine, Medical University of South Carolina, Charleston 29425, USA.. frankeae@musc.edu
SOURCE: Biochemistry, (1996 Nov 26) 35 (47) 14749-56. Journal code: 0370623. ISSN: 0006-2960.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199701
ENTRY DATE: Entered STN: 19970128
Last Updated on STN: 19970128
Entered Medline: 19970102

AB **Ricin** toxin, the heterodimeric 65 kDa glycoprotein synthesized in castor bean seeds, consists of a cell binding lectin subunit (RTB) disulfide linked to an rRNA N-glycosidase protein synthesis inactivating subunit (RTA). While X-ray crystallography and equilibrium dialysis suggested two sugar-combining sites located in subdomains 1 alpha and 2 gamma, biochemical and mutational analyses suggested the existence of a third lectin site. We performed oligonucleotide-directed mutagenesis on RTB cDNA to create mutants with modifications in subdomains 1 alpha, 2 gamma, and either 1 beta or 2 alpha. The triple-site **mutant** RTBs were expressed in insect cells. Partially purified recombinant proteins obtained from infected cell extracts and cell supernatants were characterized for asialofetuin and cell binding, immunoreactivites, ability to reassociate with RTA, and recombinant heterodimer cell cytotoxicity. Yields of both triple-site mutants were similar to the parent double-site **mutant**. Both mutants showed immunoreactivity with a panel of anti-RTB monoclonal and polyclonal antibodies. The triple-site **mutant** with modification of amino acid residues in subdomains 1 alpha, 2 alpha, and 2 gamma bound asialofetuin and cells similarly to the parent 1 alpha, 2 gamma, subdomain **mutant**. In contrast, the 1 alpha, 1 beta, 2 gamma subdomain triple-site **mutant** had a one and one-half log decrease in asialofetuin and cell binding relative to the parent double-site **mutant**. The 1 alpha, 2 alpha, 2 gamma triple-site **mutant** and 1 alpha, 2 gamma parent protein had sugar binding which was inhibited by 3-27-fold by lactose and asialofetuin. Both triple-site mutants reassociated well with RTA. The 1 alpha, 2 alpha, 2 gamma triple-site **mutant**-RTA was equally cytotoxic to mammalian cells as the double-site **mutant** -RTA heterodimer. In contrast, the 1 alpha, 1 beta, 2 gamma triple-site **mutant**-RTA was 25 times less toxic than the double **mutant** and 20 times **more toxic** than RTA alone. These data support a model for at least three lectin-binding subdomains in RTB.

L19 ANSWER 21 OF 41 MEDLINE on STN DUPLICATE 16

ACCESSION NUMBER: 95379062 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7650723
TITLE: Role of **exotoxin A** in inducing severe Pseudomonas aeruginosa infections in mice.
AUTHOR: Miyazaki S; Matsumoto T; Tateda K; Ohno A; Yamaguchi K
CORPORATE SOURCE: Department of Microbiology, Toho University School of Medicine, Tokyo, Japan.
SOURCE: Journal of medical microbiology, (1995 Sep) 43 (3) 169-75. Journal code: 0224131. ISSN: 0022-2615.
PUB. COUNTRY: SCOTLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199509
ENTRY DATE: Entered STN: 19951005
Last Updated on STN: 20020420
Entered Medline: 19950925

AB The effects of **exotoxin A** (EXA) from Pseudomonas aeruginosa on polymorphonuclear leucocytes (PMNLs) were studied in a mouse model and in vitro. P. aeruginosa PA103, which produced EXA, was 20 times **more virulent** for normal mice than was its EXA-deficient **mutant**, PA103-29. EXA was detected in the plasma of mice infected with P. **aeruginosa** PA103, and its presence correlated with increasing numbers of bacteria in the blood and internal organs. A monoclonal antibody (MAb) against EXA prevented the death of the mice if it was given simultaneously with, or 2 h before, infection with strain PA103. The number of PMNLs in murine blood decreased by 50% within 30 min of intravenous injection of EXA, but this decrease was prevented by simultaneous or prior injection of MAb to the toxin. EXA inhibited in-vitro phagocytosis and killing of P. aeruginosa by human and murine PMNLs and decreased the number of the PMNLs by between 60 and 68%. Collectively, these results not only confirm that EXA is toxic in vivo, but also suggest that this toxin accelerates the growth of virulent P. aeruginosa in mice.

L19 ANSWER 24 OF 41 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 18
ACCESSION NUMBER: 1994:180822 BIOSIS
DOCUMENT NUMBER: PREV199497193822
TITLE: An immunotoxin with increased activity and homogeneity produced by reducing the number of lysine residues in recombinant Pseudomonas exotoxin.
AUTHOR(S): Debinski, Waldemar; Pastan, Ira [Reprint author]
CORPORATE SOURCE: Lab. Molecular Biology, Div. Cancer Biology, Diagnosis Cent., National Cancer Inst., National Inst. Health, 37/4E16, 9000 Rockville Pike, Bethesda, MD 20892, USA
SOURCE: Bioconjugate Chemistry, (1994) Vol. 5, No. 1, pp. 40-46. CODEN: BCCHES. ISSN: 1043-1802.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 26 Apr 1994
Last Updated on STN: 27 Apr 1994

AB Pseudomonas **exotoxin A** (PE) is a protein composed of 613 amino acids arranged into three major, and one minor, domains. Immunotoxins (ITs) containing PE38, a **mutant** form of PE which lacks the cell binding domain (Ia, amino acids 1-252) and 16 amino acids from domain Ib (amino acids 365-380), are extremely potent cytotoxic agents which can cause a complete regression of various human carcinomas grown in nude mice. However, these ITs are a mixture of several different chemical forms since the coupling between the antibody and the toxin may occur between either the light or heavy chain of the antibody and one of the four primary amino groups present on the truncated toxin. To modify

the toxin with heterobifunctional crosslinking reagents only at specific sites, we replaced lysines 590 and 606 with glutamines and lysine 613 with arginine (PE38QQR). We also added two different peptide sequences, each containing a lysine residue, at the N-terminus of PE38. In one of these the sequence is ANLAEEAFK ("Lys" peptide), and in the other, the sequence is LQGTKLMAEE ("NLys" peptide). The **mutant** toxins were coupled using a thioether linkage to monoclonal antibody B3 which recognizes an antigen present in large amounts on many human cancers. PE38QQR-containing recombinant toxins can only be linked to an antibody through the N-terminal methionine or the lysine within the peptide. B3-LysPE38QQR and B3-NLysPE38QQR were four times **more cytotoxic** to target cells than the corresponding B3-LysPE38 and B3-NLysPE38 ITs. Furthermore, the antitumor effect of B3-NLysPE38QQR was significantly greater than that of B3-NLysPE38. We conclude that B3-LysPE38QQR and B3-NLysPE38QQR are more active because they are more homogenous components with all the antibody coupled to the N-terminus of the toxin and not some to the C-terminus, producing ITs with very low cytotoxic activity.

L19 ANSWER 25 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1994:105024 CAPLUS
DOCUMENT NUMBER: 120:105024
TITLE: **Mutant** cytokines having increased receptor affinity
INVENTOR(S): Lakkis, Fadi; Murphy, John R.
PATENT ASSIGNEE(S): University Hospital, USA
SOURCE: PCT Int. Appl., 28 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9321308	A1	19931028	WO 1993-US3613	19930416
W: AT, AU, BB, BG, BR, CA, CH, CZ, DE, DK, ES, FI, GB, HU, JP, KP, KR, LK, LU, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SK, UA, VN				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9342891	A1	19931118	AU 1993-42891	19930416
PRIORITY APPLN. INFO.:			US 1992-870500	19920417
			WO 1993-US3613	19930416

AB A variant of a naturally-occurring cytokine has a neutral amino acid substituted for a neg.-charged amino acid within 2 amino acids immediately upstream or downstream from a Phe-Leu or Tyr-Leu sequence in a helical domain. The variant cytokine has an increased affinity for the receptor. A hybrid mol. comprises a receptor-binding portion of the variant cytokine joined together covalently with a mol. having enzymic activity (e.g., a cytotoxin). The hybrid mol. decreases cell viability. DAB389-mIL-4, a fusion protein containing **diphtheria toxin** having a deletion of 97 amino acids (Thr387-His485; the generalized cell binding domain) replaced with murine IL-4, was altered by site-directed and in-frame deletion mutagenesis to alter the mIL-4 portion of DAB389-mIL-4. Deletion of the C-terminal 15 amino acids of mIL-4; substitution of Phe496 with Pro, Ala, or Tyr; or substitution of Leu497 with Ala or Glu decreased binding to the mIL-4 receptor and cytotoxicity. In contrast, the substitution of the neg.-charged residue Asp495 with Asn resulted in a 4-fold **increase** in **cytotoxic** potency and binding affinity to mIL-4 receptor bearing cells in vitro.

L19 ANSWER 26 OF 41 MEDLINE on STN DUPLICATE 19
ACCESSION NUMBER: 93186791 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8444864
TITLE: Heparin-binding transforming growth factor alpha-Pseudomonas **exotoxin A.** A heparan sulfate-modulated recombinant toxin cytotoxic to cancer cells and proliferating smooth muscle cells.
AUTHOR: Mesri E A; Kreitman R J; Fu Y M; Epstein S E; Pastan I
CORPORATE SOURCE: Division of Cancer Diagnosis Biology and Centers, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20892.
SOURCE: Journal of biological chemistry, (1993 Mar 5) 268 (7) 4853-62.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199304
ENTRY DATE: Entered STN: 19930416
Last Updated on STN: 20000303
Entered Medline: 19930406

AB TGF alpha-PE40, a recombinant toxin in which transforming growth factor alpha (TGF alpha) is fused to a **mutant** form of Pseudomonas exotoxin, is selectively cytotoxic to cells bearing epidermal growth factor (EGF) receptors. Heparin binding EGF-like growth factor is a potent mitogen for smooth muscle cells capable of binding to both the EGF receptor and to immobilized heparin (Higashiyama, S., Abraham, J., Miller, J., Fiddes, J., and Klagsbrun, M. (1991) Science 251, 936-938). To study the effect of the heparin-binding domain in a chimeric toxin targeted to the EGF receptor, we fused the DNA sequence corresponding to the putative NH2-terminal heparin-binding (HB) domain of HB-EGF to chimeric toxins composed of TGF alpha and two different recombinant forms of Pseudomonas exotoxin (PE). One of these is a truncated form of PE devoid of the binding domain (TGF alpha-PE38); another is a **mutant** form of full-length toxin containing inactivating mutations in the binding domain and an altered carboxyl terminus (TGF alpha-PE4EKDEL). The resulting chimeric toxins HB-TGF alpha-PE38 and HB-TGF alpha-PE4EKDEL were expressed in Escherichia coli as inclusion bodies, refolded, and purified by heparin affinity chromatography. Both of the toxins were eluted from heparin at 0.8 M NaCl, in contrast to their respective TGF alpha toxins which were eluted at 0.15 M. Binding studies on A431 cells showed that the HB-TGF alpha toxins bound to the EGF receptor with an affinity similar to that of the TGF alpha toxins. However, cell killing studies on a panel of malignant cell lines showed that cytotoxicity was strongly affected by the presence of the HB domain. Cell lines expressing high numbers of EGF receptors such as A431 and KB were less sensitive to toxins containing the HB domain. Cells with low number of EGF receptors had similar responses to both types of toxins (MCF-7 and LNCaP) or were more sensitive to the toxin with the added HB domain (HEP-G2). HB-TGF alpha-PE4EKDEL was over 10-fold **more cytotoxic** against proliferating vascular smooth muscle cells (VSMC) than to quiescent VSMC. Moreover, HB-TGF alpha-PE4EKDEL was 6-fold more potent than TGF alpha-PE4EKDEL to proliferating VSMC. Competition studies with EGF and/or heparin showed that heparin blocks the cytotoxicity of HB-TGF toxins and the inhibitory action of heparin is stronger in cells expressing lower number of EGF receptors.(ABSTRACT TRUNCATED AT 400 WORDS)

L19 ANSWER 27 OF 41 MEDLINE on STN DUPLICATE 20
ACCESSION NUMBER: 93054682 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1429683
TITLE: Alanine scanning mutagenesis identifies surface amino acids

on domain II of Pseudomonas exotoxin required for cytotoxicity, proper folding, and secretion into periplasm.
AUTHOR: Kasturi S; Kihara A; FitzGerald D; Pastan I
CORPORATE SOURCE: Laboratory of Molecular Biology, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20892.
SOURCE: Journal of biological chemistry, (1992 Nov 15) 267 (32) 23427-33.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199212
ENTRY DATE: Entered STN: 19930122
Last Updated on STN: 20020420
Entered Medline: 19921216

AB Pseudomonas **exotoxin A** (PE) is a single polypeptide chain that contains 613 amino acids and is arranged into three major structural domains. Domain Ia is responsible for cell recognition, domain II for translocation of PE across the membrane, and domain III for ADP-ribosylation of elongation factor 2. Recombinant PE can be produced in Escherichia coli and is efficiently secreted into the periplasm when an OmpA signal sequence is present. To investigate the role of the amino acids located on the surface of domain II in the action of the toxin against mammalian cells, we substituted alanine for each of the 27 surface amino acids present in domain II. Surprisingly, all 27 **mutant** proteins had some alteration in cytotoxicity when tested on human A431 or MCF7 cells or mouse L929 cells. Native PE has a compact structure and therefore is relatively protease resistant and very little ADP-ribosylation activity is detected in the absence of the denaturing agents like urea and dithiothreitol. Several of the mutations resulted in altered protease sensitivity of the toxin. Seven of the **mutant** molecules exhibited ADP-ribosylation activity without urea and dithiothreitol, indicating they are partially unfolded. Out of these seven mutants, six had **increased cytotoxic** activity on at least one of the target cell lines and the other retained its native cytotoxic potency.

L19 ANSWER 28 OF 41 MEDLINE on STN DUPLICATE 21
ACCESSION NUMBER: 92380987 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1512230
TITLE: A recombinant form of Pseudomonas exotoxin directed at the epidermal growth factor receptor that is cytotoxic without requiring proteolytic processing.
AUTHOR: Theuer C P; FitzGerald D; Pastan I
CORPORATE SOURCE: Laboratory of Molecular Biology, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20892.
SOURCE: Journal of biological chemistry, (1992 Aug 25) 267 (24) 16872-7.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199209
ENTRY DATE: Entered STN: 19921018
Last Updated on STN: 20020420
Entered Medline: 19920925

AB Pseudomonas **exotoxin A** is composed of three structural domains that mediate cell recognition (I), membrane translocation (II), and ADP-ribosylation (III). Within the cell, the toxin is cleaved within domain II to produce a 37-kDa carboxyl-terminal fragment, containing amino

acids 280-613, which is translocated to the cytosol and causes cell death. In this study, we constructed a **mutant** protein (PE37), composed of amino acids 280-613 of Pseudomonas **exotoxin A**, which does not require proteolysis to translocate. PE37 was targeted specifically to cells with epidermal growth factor receptors by inserting transforming growth factor-alpha (TGF-alpha) after amino acid 607 near the carboxyl terminus of Pseudomonas **exotoxin A**. PE37/TGF-alpha was very cytotoxic to cells with epidermal growth factor receptors. It was severalfold **more cytotoxic** than a derivative of full-length Pseudomonas **exotoxin A** containing TGF-alpha in the same position, probably because the latter requires intracellular proteolytic processing to exhibit its cytotoxicity, and proteolytic processing is not 100% efficient. Deletion of 2, 4, or 7 amino acids from the amino terminus of PE37/TGF-alpha **greatly** diminished **cytotoxic** activity, indicating the need for a proper amino-terminal sequence. In addition, a **mutant** containing an internal deletion of amino acids 314-380 was minimally active, indicating that other regions of domain II are also required for the cytotoxic activity of Pseudomonas **exotoxin A**.

L19 ANSWER 29 OF 41 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 92282042 EMBASE
DOCUMENT NUMBER: 1992282042
TITLE: Interactions of **diphtheria toxin** B-fragment with cells. Role of amino- and carboxyl-terminal regions.
AUTHOR: Stenmark H.; Ariansen S.; Afanasiev B.N.; Olsnes S.
CORPORATE SOURCE: Institute for Cancer Research, Norwegian Radium Hospital,Montebello, Oslo 3, Norway
SOURCE: Journal of Biological Chemistry, (1992) 267/13 (8957-8962). ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
052 Toxicology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The B-fragment of **diphtheria toxin** binds to cell surface receptors and facilitates entry of the enzymatically active A-fragment into the cytosol. The roles of the amino- and carboxyl-terminal regions of the B-fragment in interactions with the cell membrane were studied by measuring specific binding, insertion into membranes at low pH, and formation of cation- selective channels, as well as by toxicity measurements after association with active A-fragment. Deletion of the amino-terminal 12 amino acids of the B-fragment did not affect its ability to bind to receptors and to form ion channels at low pH, whereas both abilities were strongly impaired when one more amino acid (Trp206) was removed. Replacement of the amino-terminal 31 residues with an amphipathic sequence from human apolipoprotein A1 restored receptor binding but not ion channel formation. The binding to cells was virtually abolished when 9 residues were deleted from the carboxyl terminus. Deletion of only 4 residues or extension by 12 residues did not prevent specific binding, but reduced insertion, channel formation, and toxicity. Those deletions that reduced receptor binding ability increased the trypsin sensitivity of the B-fragment. The results indicate that the amino- and carboxyl-terminal regions of **diphtheria toxin** B-fragment are important for receptor binding, possibly because they contribute to keep the B-fragment in a binding-competent conformation. Small alterations in the carboxyl-terminal end reduced insertion, channel formation, and **toxicity more** than the ability of the B-fragment to bind to cells.

L19 ANSWER 30 OF 41 MEDLINE on STN DUPLICATE 22
ACCESSION NUMBER: 93028322 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1409544
TITLE: Phe496 and Leu497 are essential for receptor binding and cytotoxic action of the murine interleukin-4 receptor targeted fusion toxin DAB389-mIL-4.
AUTHOR: Lakkis F; Landgraf B; Wen Z; Strom T B; Murphy J R
CORPORATE SOURCE: Evans Department of Clinical Research, University Hospital, Boston, MA 02118.
CONTRACT NUMBER: U01 CA-48626 (NCI)
SOURCE: Protein engineering, (1992 Apr) 5 (3) 241-8.
Journal code: 8801484. ISSN: 0269-2139.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199211
ENTRY DATE: Entered STN: 19930122
Last Updated on STN: 19980206
Entered Medline: 19921103

AB DAB389-mIL-4 is a murine interleukin-4 (mIL-4) **diphtheria toxin**-related fusion protein which has been shown to be selectively toxic to cells expressing the mIL-4 receptor. In this report, we have used site-directed and in-frame deletion mutagenesis to study the role of the putative C-terminal alpha-helix (helix E) of the mIL-4 component of DAB389-mIL-4 in the intoxication process. We demonstrate that deletion of the C-terminal 15 amino acids of the fusion toxin leads to loss of cytotoxicity. The substitution of Phe496 with either Pro, Ala or Tyr, results in a greater than 20-fold decrease in cytotoxic activity of the respective **mutant** fusion toxins. In addition, substitution of Leu497 with either Ala or Glu results in a similar loss of cytotoxic activity. All of these **mutant** forms of the mIL-4 fusion toxin demonstrate a significant decrease in binding affinity (Ki) to the mIL-4 receptor in a competitive radioligand binding assay. In marked contrast, however, the substitution of Asp495 with Asn results in a 4-fold **increase** in **cytotoxic** potency and binding affinity to mIL-4 receptor bearing cells in vitro.

L19 ANSWER 31 OF 41 MEDLINE on STN DUPLICATE 23
ACCESSION NUMBER: 92062073 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1953725
TITLE: A proper amino terminus of **diphtheria toxin** is important for cytotoxicity.
AUTHOR: Chaudhary V K; FitzGerald D J; Pastan I
CORPORATE SOURCE: Laboratory of Molecular Biology, National Cancer Institute, National Institutes of Health, Bethesda, MD 20892.
SOURCE: Biochemical and biophysical research communications, (1991 Oct 31) 180 (2) 545-51.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199112
ENTRY DATE: Entered STN: 19920124
Last Updated on STN: 19920124
Entered Medline: 19911202

AB A series of deletions and substitutions were made at the 5' end of the gene fusion between the first 388 codons of **diphtheria toxin** (DT) and a cDNA encoding human IL2. The chimeric protein (DT388-IL2) was expressed and purified from E. coli and found to be very

cytotoxic to a human T cell line, HUT 102, that expresses a large number of IL2 receptors. Deletion of the first five amino acids of DT resulted in a non-cytotoxic chimeric protein that had both ADP-ribosylation activity and IL2 receptor binding activity. Deletion of the first two amino acids of DT had little effect on cytotoxicity, while deletion of the first four amino acids or of two acidic residues at positions 3 and 4 **greatly** reduced **cytotoxicity**. Unexpectedly, a **mutant** containing a single leucine in place of the first two amino acids (gly, ala) was 2-3 fold more active. The amino terminus of DT may participate in the translocation of the A chain to the cytosol in a manner similar to Pseudomonas exotoxin (PE) in which a specific C-terminal sequence has been proposed to be involved in its cytotoxicity.

L19 ANSWER 32 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:650006 CAPLUS
DOCUMENT NUMBER: 115:250006
TITLE: Enhancement of the cytotoxicity of mistletoe lectin-1 (ML-1) by high pH or perturbation in Golgi functions
AUTHOR(S): Yoshida, T.; Zhang, M.; Chen, C.; Franz, H.; Wu, H. C.
CORPORATE SOURCE: Dep. Microbiol., Unif. Serv. Univ. Health Sci., Bethesda, MD, 20814-4799, USA
SOURCE: Pharmazie (1991), 46(5), 349-51
CODEN: PHARAT; ISSN: 0031-7144
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The cytotoxicity of ML-1 in CHO and Vero cells is enhanced by increasing the endosomal pH ammonium chloride treatment or **mutant** with defective endosomal acidification or by perturbation in Golgi functions (nigericin treatment or **mutant** altered in Golgi functions). In both aspects, ML-1 resembles closely **ricin** in the **cytotoxic** process: a **more** alkaline pH in endosomal vesicles favors the release of ML-1 and **ricin** and the involvement of Golgi region as a potential site of toxin release into the cytosol. Further studies on the intracellular trafficking and the translocation process of these toxins are needed to elucidate the mechanisms of toxin release in sensitive host cells.

L19 ANSWER 33 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:96250 CAPLUS
DOCUMENT NUMBER: 114:96250
TITLE: Cytotoxic recombinant Pseudomonas endotoxin and target-specific fusion products
INVENTOR(S): Pastan, I.
PATENT ASSIGNEE(S): National Institutes of Health, USA
SOURCE: U. S. Pat. Appl., 33 pp. Avail. NTIS Order No. PAT-APPL-7-759 635.
CODEN: XAXXAV
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 459635	A0	19900415	US 1990-459635	19900102
US 522563	A0	19910515	US 1990-522563	19900514
US 5458878	A	19951017		
CA 2072891	AA	19910703	CA 1990-2072891	19901227
CA 2072891	C	19991221		
WO 9109949	A1	19910711	WO 1990-US7421	19901227
W: AU, CA, JP				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE				

AU 9172424	A1	19910724	AU 1991-72424	19901227
AU 644139	B2	19931202		
EP 509056	A1	19921021	EP 1991-904103	19901227
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
JP 05502032	T2	19930415	JP 1991-504333	19911217
US 5705163	A	19980106	US 1995-461233	19950605
PRIORITY APPLN. INFO.:			US 1990-459635	19900102
			US 1990-522563	A3 19900514
			WO 1990-US7421	A 19901227

AB The carboxyl terminus of Pseudomonas **exotoxin A** (PE), residues Arg609-Lys613, dets. the cytotoxic activity of the exotoxin. Peptide sequence Lys-Asp-Glu-Leu (KDEL), which is responsible for retaining newly formed proteins within the endoplasmic reticulum, has similar biol. function to the carboxyl terminus of PE. When KDEL is fused to a carboxyl terminus-deleted PE **mutant** (non-cytotoxic), it restored the cytotoxic activity of the toxin. A recognition mol. such as antibody may be fused to the carboxyl terminus of PE to increase the potency of the chimeric toxin. Fusion proteins of PE and transforming growth factor α were prepared, and their cytotoxic activity against Swiss 3T3 cells determined The fusion proteins with active carboxyl terminus were ≥50 fold **more cytotoxic** than that containing inactive PE carboxyl terminus.

L19 ANSWER 34 OF 41 MEDLINE on STN DUPLICATE 24

ACCESSION NUMBER:	90353427 MEDLINE
DOCUMENT NUMBER:	PubMed ID: 2117543
TITLE:	**Increased cytotoxicity** of **ricin** in a putative Golgi-defective **mutant** of Chinese hamster ovary cell.
AUTHOR:	Yoshida T; Chen C H; Zhang M S; Wu H C
CORPORATE SOURCE:	Department of Microbiology, Uniformed Services University of the Health Sciences, Bethesda, Maryland 20814-4799.
CONTRACT NUMBER:	GM28810 (NIGMS)
SOURCE:	Experimental cell research, (1990 Sep) 190 (1) 11-6. Journal code: 0373226. ISSN: 0014-4827.
PUB. COUNTRY:	United States
DOCUMENT TYPE:	Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:	English
FILE SEGMENT:	Priority Journals
ENTRY MONTH:	199009
ENTRY DATE:	Entered STN: 19901026 Last Updated on STN: 19990129 Entered Medline: 19900921

AB We have studied the cytotoxicity of **ricin** in a monensin-resistant **mutant** (MonR-31) of Chinese hamster ovary (CHO) cell line which is presumably altered in Golgi functions/structures. The **cytotoxicity** of **ricin** was **increased** in MonR-31 **mutant** cells compared with that in its parental CHO cells. In wild-type CHO cells, the cytotoxicity of **ricin** was enhanced by HN4Cl, bafilomycin A1, or nigericin. The enhancement of **ricin** cytotoxicity by these compounds was greatly reduced in MonR-31 **mutant** cells. Brefeldin A (BFA), which disrupts the structure of the Golgi apparatus, inhibits the cytotoxicity of **ricin** in both CHO and MonR-31 cells. We have also examined the effects of glycosylation inhibitors and the removal of high mannose oligosaccharide chains in **ricin** on the **ricin** hypersensitivity in MonR-31 cells. The hypersensitivity of MonR-31 cells to **ricin** is apparently not due to any difference in glycosylation between CHO and MonR-31 cells or in the processing of oligosaccharides on **ricin** by the target cells. Nigericin at low concentration (10 nM), which has no effect on the cytotoxicity of **diphtheria toxin**, enhances the **ricin**

cytotoxicity, but inhibits the **modeccin** cytotoxicity. Our results suggest that important step(s) in the intoxication process of CHO cells by **ricin** and **modeccin** take place in the Golgi region.

L19 ANSWER 35 OF 41 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
DUPLICATE 25

ACCESSION NUMBER: 1989:494307 BIOSIS
DOCUMENT NUMBER: PREV198988120844; BA88:120844
TITLE: DOMAIN II MUTANTS OF PSEUDOMONAS EXOTOXIN DEFICIENT IN TRANSLOCATION.
AUTHOR(S): JINNO Y [Reprint author]; OGATA M; CHAUDHARY V K; WILLINGHAM M C; ADHYA S; FITZGERALD D; PASTAN I
CORPORATE SOURCE: LAB MOL BIOL, NATL CANCER INST, NATL INST HEALTH, BETHESDA, MARYLAND 20892, USA
SOURCE: Journal of Biological Chemistry, (1989) Vol. 264, No. 27, pp. 15953-15959.
CODEN: JBCHA3. ISSN: 0021-9258.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 2 Nov 1989
Last Updated on STN: 4 Nov 1989

AB Pseudomonas exotoxin (PE) kills mammalian cells in a complex process that involves cell surface binding, internalization by endocytosis, translocation to the cytosol, and ADP-ribosylation of elongation factor 2. PE is a three-domain protein in which domain I binds to the cell surface, domain II promotes translocation into the cytosol, and domain III carries out ADP-ribosylation. To determine how translocation occurs, we have **mutated** all the arginine residues in domain II and found that mutations at positions 276 and 279 **greatly** diminished the **cytotoxicity** of PE and mutations 330 and 337 substantially reduced cytotoxicity. Biochemical studies indicate that after internalization into an endocytic compartment, the PE molecule undergoes a specific and saturable intracellular interaction and this interaction is deficient in an Arg276 → Gly **mutant**. Our data suggest that the translocation process of PE involves a specific interaction of Arg276 (and possibly Arg279, Arg330, and Arg337) with components of an intracellular compartment.

L19 ANSWER 36 OF 41 MEDLINE on STN
ACCESSION NUMBER: 88198152 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3129420
TITLE: Restoration of enzymic activity and cytotoxicity of **mutant**, E553C, Pseudomonas aeruginosa **exotoxin A** by reaction with iodoacetic acid.
AUTHOR: Lukac M; Collier R J
CORPORATE SOURCE: Department of Microbiology and Molecular Genetics, Harvard Medical School, Boston, Massachusetts.
CONTRACT NUMBER: AI-22021 (NIAID)
AI-22848 (NIAID)
SOURCE: Journal of biological chemistry, (1988 May 5) 263 (13) 6146-9.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198806
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 20020420

Entered Medline: 19880606

AB Pseudomonas aeruginosa **exotoxin A** (ETA) is inactivated greater than 1,000-fold when an active site glutamic acid, E553, is **mutated** to aspartic acid (Douglas, C.M., and Collier, R. J. (1987) J. Bacteriol. 169, 4967-4971). To test the effect of creating a carboxyl-containing side chain at position 553 longer than that of glutamic acid, we first replaced Glu-553 with cysteine by site-directed mutagenesis of cloned ETA and then carboxymethylated the cysteine side chain with iodoacetic acid. The E553C mutation reduced ADP-ribosyltransferase and **cytotoxic** activities **greater** than 10,000-fold. Reaction of the **mutant** with iodoacetic acid enhanced enzymic activity 2,500-fold, to a level approximately one-sixth that of wild type toxin, and restored cytotoxicity to a slightly lesser extent. Iodoacetamide did not activate the **mutant**, and neither iodoacetic acid nor iodoacetamide affected the activity of wild type toxin. These results show that the carboxyl group of Glu-553 is important for ADP-ribosylation activity and imply flexibility in the enzyme-substrate complex in accommodating the slightly longer S-carboxymethylcysteine side chain. This general approach may have applications in protein engineering as well as in studying carboxyl side chain functions in enzymes.

L19 ANSWER 38 OF 41 MEDLINE on STN DUPLICATE 26
ACCESSION NUMBER: 86215055 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3085652
TITLE: Properties of baby-hamster kidney (BHK) cells treated with Swainsonine, an inhibitor of glycoprotein processing. Comparison with **ricin**-resistant BHK-cell mutants.
AUTHOR: Foddy L; Feeney J; Hughes R C
SOURCE: Biochemical journal, (1986 Feb 1) 233 (3) 697-706. Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198605
ENTRY DATE: Entered STN: 19900321
Last Updated on STN: 19900321
Entered Medline: 19860530

AB Baby-hamster kidney (BHK) cells were grown continuously in long-term monolayer culture in the presence of Swainsonine, an inhibitor of alpha-mannosidase II, a processing enzyme involved in glycoprotein biosynthesis. The asparagine-linked oligosaccharides (N-glycans) were isolated from Pronase-digested cells by gel filtration, ion-exchange chromatography and affinity chromatography on concanavalin A--Sepharose and lentil lectin--Sepharose. The major N-glycans, analysed by 500 MHz 1H-n.m.r. spectroscopy, were identified as hybrid structures containing five mannose residues and neutral high-mannose N-glycans. The major hybrid species contained a core-substituted fucose alpha(1----6) residue and a NeuNAc alpha(2----3)Gal beta(1----4)GlcNAc terminal sequence; smaller amounts of non-sialylated and non-fucosylated hybrid structures were also detected. Swainsonine-treated cells also produced neutral oligosaccharides containing a single reducing N-acetylglucosamine residue substituted with polymannose sequences. The glycopeptide composition of Swainsonine-treated BHK cells resembles closely that of the **ricin** -resistant BHK cell **mutant**, RicR21 [P. A. Gleeson, J. Feeney and R. C. Hughes (1985) Biochemistry 24, 493-503], except the hybrid structures of RicR21 cells contain three, not five, mannose residues. Like RicR21 cells, Swainsonine-treated BHK cells showed a greatly **increased** resistance to **ricin cytotoxicity**, but not to **modeccin**, another galactose-binding lectin. These effects were readily reversed on removal of Swainsonine and growth in

normal medium.

L19 ANSWER 40 OF 41 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1978:401445 CAPLUS
DOCUMENT NUMBER: 89:1445
TITLE: Formation of a hybrid toxin from **ricin** agglutinin and a non-toxic **mutant** protein of **diphtheria toxin**
AUTHOR(S): Uchida, Tsuyoshi; Yamaizumi, Masaru; Okada, Yoshio
CORPORATE SOURCE: Res. Inst. Microb. Dis., Osaka Univ., Suita, Japan
SOURCE: Biochemical and Biophysical Research Communications (1978), 81(2), 268-73
CODEN: BBRCA9; ISSN: 0006-291X
DOCUMENT TYPE: Journal
LANGUAGE: English
AB CRM45, a non-toxic **mutant** protein of **diphtheria toxin**, was treated with glutaraldehyde and conjugated to **ricin** agglutinin. The hybrid protein thus obtained was purified by gel filtration and affinity chromatog. The purified hybrid toxin was .apprx.8-10 times **more toxic** than **ricin** agglutinin when tested in mice and cultured L cells.

L19 ANSWER 41 OF 41 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 78123550 EMBASE
DOCUMENT NUMBER: 1978123550
TITLE: Ricinus communis toxin mediated inhibition of protein synthesis in cell free extracts of a toxin resistant variant mouse lymphoma cell line.
AUTHOR: Robbins J.C.; Hunter T.R.; Nicolson G.L.
CORPORATE SOURCE: Dept. Cancer Biol., Salk Inst. Biol. Studies, San Diego, Calif. 92112, United States
SOURCE: Journal of Supramolecular and Cellular Biochemistry, (1977) 5/4 (515-520).
CODEN: JSPMAW
COUNTRY: United States
DOCUMENT TYPE: Journal
FILE SEGMENT: 037 Drug Literature Index
029 Clinical Biochemistry
005 General Pathology and Pathological Anatomy
030 Pharmacology
LANGUAGE: English
AB Ricinus communis agglutinin II (RCAII, **ricin**, toxin) at low concentrations inhibits protein synthesis in cell-free extracts, but not in intact cells, of an RCAII-resistant mouse lymphoma variant cell line. The concentration dependence of the inhibition by RACII was the same in cell-free extracts of both RCAII-resistant variant and RACII-sensitive parental cells, while intact parental cells are 250 times **more** sensitive to RCAII **toxicity**. The onset of RCAII inhibition of cell-free protein synthesis was extremely rapid in both cases, being complete in a few minutes. Under these conditions RCAII inhibits protein synthesis in intact RCAII-sensitive parental cells, but maximal inhibition requires several hr to occur. These results support previous electron microscopic observations that the variant cells are defective in the uptake of RCAII by endocytosis at low toxin concentrations.

=> FIL STNGUIDE
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	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	77.17	347.19

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                                                     ENTRY    SESSION
CA SUBSCRIBER PRICE                                  -3.50      -7.70

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LAST RELOADED: Nov 12, 2004 (20041112/UP).

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                                                     ENTRY    SESSION
FULL ESTIMATED COST                                   1.50     348.69

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CA SUBSCRIBER PRICE                                   0.00      -7.70

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     (FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18
     NOV 2004
L1             0 SHIGA 5A TOXIN
L2           193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3             1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4         20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5           182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6           122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7            16 L2 AND L6
L8            10 DUP REM L7 (6 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18
     NOV 2004
L9             2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
L10            2 DUP REM L9 (0 DUPLICATES REMOVED)
L11            1 L2 AND L4
L12          125 DUP REM L2 (68 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 16:42:37 ON 18 NOV 2004
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      FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:58:30 ON 18
      NOV 2004
L13         2725 (MUTATED OR MUTANT) AND ((VIRULENT OR VIRULENCE OR TOXICITY OR
L14           15 L13 AND SHIGA (3N) TOXIN
L15            6 DUP REM L14 (9 DUPLICATES REMOVED)

      FILE 'STNGUIDE' ENTERED AT 17:03:25 ON 18 NOV 2004
L16            0 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (

      FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 17:19:55 ON 18
      NOV 2004
L17        32942 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (
L18          108 L13 AND L17
L19           41 DUP REM L18 (67 DUPLICATES REMOVED)

      FILE 'STNGUIDE' ENTERED AT 17:31:53 ON 18 NOV 2004

      FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 17:46:35 ON 18
      NOV 2004

=> (Shiga (w) toxin or Shiga (w)like(w)toxin) and l13
L20           15 (SHIGA (W) TOXIN OR SHIGA (W) LIKE(W) TOXIN) AND L13

=> l20 not l14
L21            0 L20 NOT L14

=> l13 and (Gariepy J?/au or Bray M?/au)
L22            0 L13 AND (GARIEPY J?/AU OR BRAY M?/AU)

=> (Gariepy J?/au or Bray M?/au)
L23         1647 (GARIEPY J?/AU OR BRAY M?/AU)

=> e Griepy Jean/au
E1             1     GRIEPP RANDALL G/AU
E2             1     GRIEPSMA J/AU
E3             0 --> GRIEPY JEAN/AU
E4             1     GRIEPY MANON/AU
E5             8     GRIER A/AU
E6             1     GRIER A G/AU
E7            17     GRIER A H/AU
E8             1     GRIER A J/AU
E9             5     GRIER A M/AU
E10            4     GRIER A R/AU
E11            1     GRIER A W/AU
E12            1     GRIER ALEXANDER R/AU

=> e Gariepy Jean/au
E1            50     GARIEPY J L/AU
E2             1     GARIEPY JASON/AU
E3           115 --> GARIEPY JEAN/AU
E4            22     GARIEPY JEAN LOUIS/AU
E5            53     GARIEPY JEROME/AU
E6             4     GARIEPY K/AU
E7             8     GARIEPY K C/AU
E8             3     GARIEPY KATHLEEN C/AU
E9             3     GARIEPY KATHLEEN COX/AU
E10           32     GARIEPY L/AU
E11            2     GARIEPY L B/AU
E12            1     GARIEPY L E/AU

=> s e3 or e4
```

```
L24         137 "GARIEPY JEAN"/AU OR "GARIEPY JEAN LOUIS"/AU

=> s e1 or e3 or e4
L25         187 "GARIEPY J L"/AU OR "GARIEPY JEAN"/AU OR "GARIEPY JEAN LOUIS"/AU

=> e Bray M/au
E1            1      BRAY LOUIS/AU
E2            2      BRAY LOUISE/AU
E3          270 --> BRAY M/AU
E4          227      BRAY M A/AU
E5            1      BRAY M B/AU
E6            5      BRAY M C/AU
E7            1      BRAY M D/AU
E8            4      BRAY M E/AU
E9            1      BRAY M F/AU
E10           1      BRAY M I/AU
E11          34      BRAY M J/AU
E12           5      BRAY M K/AU

=> e Bray Mark/au
E1            2      BRAY MARGARET M/AU
E2            2      BRAY MARI V/AU
E3            0 --> BRAY MARK/AU
E4           14      BRAY MARK ANTHONY/AU
E5            1      BRAY MARK ANTHONY PETER/AU
E6            1      BRAY MARK E/AU
E7           33      BRAY MARK R/AU
E8            2      BRAY MARK ROBERT/AU
E9           42      BRAY MARK W/AU
E10           5      BRAY MARTIN/AU
E11           2      BRAY MARTIN J/AU
E12           1      BRAY MARTY/AU

=> s e7 or e8
L26          35 "BRAY MARK R"/AU OR "BRAY MARK ROBERT"/AU

=> s l25 or l26
L27         217 L25 OR L26

=> (Shiga (w) toxin or Shiga (w)like(w)toxin) and l27
L28          21 (SHIGA (W) TOXIN OR SHIGA (W) LIKE(W) TOXIN) AND L27

=> dup rem l28
PROCESSING COMPLETED FOR L28
L29           16 DUP REM L28 (5 DUPLICATES REMOVED)

=> t ti l29 1-16

L29  ANSWER 1 OF 16  BIOSIS  COPYRIGHT (c) 2004 The Thomson Corporation.  on
     STN                                                    DUPLICATE 1
TI   Method for selectively purging CD77+ cells from bone marrow or peripheral
     blood.

L29  ANSWER 2 OF 16  BIOSIS  COPYRIGHT (c) 2004 The Thomson Corporation.  on
     STN
TI   Shiga-like toxins as targeted anticancer agents.

L29  ANSWER 3 OF 16  CAPLUS  COPYRIGHT 2004 ACS on STN
TI   Shiga-like toxins as targeted anticancer agents

L29  ANSWER 4 OF 16  BIOSIS  COPYRIGHT (c) 2004 The Thomson Corporation.  on
     STN
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TI Probing the surface of eukaryotic cells using combinatorial toxin libraries.

L29 ANSWER 5 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI The use of **Shiga-like toxin** 1 in cancer therapy.

L29 ANSWER 6 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Intracellular trafficking of **Shiga-like Toxin** I.

L29 ANSWER 7 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI **Shiga-like toxin** 1 as a potent purging agent for autologous stem cell grafts of patients with breast cancer, lymphoma and multiple myeloma.

L29 ANSWER 8 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
TI Cytotoxic heteromeric protein combinatorial libraries

L29 ANSWER 9 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
TI Deletion analysis of the **Shiga-like Toxin** I A chain: Minimum domain for catalytic activity and cytotoxicity.

L29 ANSWER 10 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
TI A method for selectively purging CD77+ cells from bone marrows

L29 ANSWER 11 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 2
TI Orientation in lipid bilayers of a synthetic peptide representing the C-terminus of the A1 domain of **Shiga toxin**. A polarized ATR-FTIR study.

L29 ANSWER 12 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 3
TI Insertion and orientation of a synthetic peptide representing the C-terminus of the A-1 domain of **Shiga toxin** into phospholipid membranes.

L29 ANSWER 13 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 4
TI **Shiga-like toxin** purges human lymphoma from bone marrow of severe combined immunodeficient mice.

L29 ANSWER 14 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 5
TI Local conformational change in the B-subunit of **Shiga-like toxin** 1 at endosomal pH.

L29 ANSWER 15 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
TI Serological responses to the B subunit of **Shiga-like toxin** 1 and its peptide fragments indicate that the B subunit is a vaccine candidate to counter the action of the toxin

L29 ANSWER 16 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
TI Characterization of **Shiga-like toxin** I B subunit purified from overproducing clones of the SLT-I B cistron

=> d ibib abs l29 1-16

L29 ANSWER 1 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 1

ACCESSION NUMBER: 2002:197978 BIOSIS
DOCUMENT NUMBER: PREV200200197978
TITLE: Method for selectively purging CD77+ cells from bone marrow or peripheral blood.
AUTHOR(S): **Gariepy, Jean** [Inventor, Reprint author]
CORPORATE SOURCE: 43 Chester Avenue, Toronto, Ontario, Canada
PATENT INFORMATION: US 6348446 February 19, 2002
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Feb. 19, 2002) Vol. 1255, No. 3. http://www.uspto.gov/web/menu/patdata.html. e-file. CODEN: OGUPE7. ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
ENTRY DATE: Entered STN: 13 Mar 2002
Last Updated on STN: 13 Mar 2002

AB A method for the selective purging ex vivo of CD77 positive cells from bone marrow or peripheral blood containing stem cells prior to autologous transplantation is described. The method involves treating the bone marrow or blood sample with **shiga toxin** or **shiga-like toxin**-1 to kill CD77+ cells or to remove them by affinity chromatography. The toxin selectively binds to CD77+ cells and not to other stem cells. The method offers a means for curing non-Hodgkin's lymphomas, myelomas and breast cancers expressing CD77.

L29 ANSWER 2 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

ACCESSION NUMBER: 2002:355051 BIOSIS
DOCUMENT NUMBER: PREV200200355051
TITLE: Shiga-like toxins as targeted anticancer agents.
AUTHOR(S): LaPointe, Paul [Reprint author]; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Department of Medical Biophysics, University of Toronto and The Ontario Cancer Institute, Princess Margaret Hospital, University Health Network, Toronto, Canada
SOURCE: Page, Michel [Editor]. (2002) pp. 307-318. Cancer Drug Discovery and Development. Tumor targeting in cancer therapy. print.
Publisher: Humana Press Inc., 999 Riverview Drive, Suite 208, Totowa, NJ, 07512, USA.
ISBN: 0-89603-919-6 (cloth).
DOCUMENT TYPE: Book
Book; (Book Chapter)
LANGUAGE: English
ENTRY DATE: Entered STN: 26 Jun 2002
Last Updated on STN: 26 Jun 2002

L29 ANSWER 3 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 2002:350835 CAPLUS
DOCUMENT NUMBER: 137:362218
TITLE: Shiga-like toxins as targeted anticancer agents
AUTHOR(S): LaPointe, Paul; **Gariepy, Jean**
CORPORATE SOURCE: Department of Medical Biophysics, University of Toronto and The Ontario Cancer Institute, Princess Margaret Hospital, University Health Network, Toronto, Can.
SOURCE: Tumor Targeting in Cancer Therapy (2002), 307-318. Editor(s): Page, Michel. Humana Press Inc.: Totowa,

N. J.
CODEN: 69COIT; ISBN: 0-89603-919-6
DOCUMENT TYPE: Conference; General Review
LANGUAGE: English
AB A review exploring the use of Shiga (ShT) and Shiga-like toxins (SLT) as chemotherapeutic agents in view of their receptor specificity and cytotoxicity. Specific topics discussed include intracellular routing of SLT-1, SLT-1 in cancer therapy, SLT-1 as a purging agent, SLT-1 as an in vivo therapeutic agent, the role of SLT-1 in treating patients with brain cancer, and future directions.
REFERENCE COUNT: 64 THERE ARE 64 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L29 ANSWER 4 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2001:302761 BIOSIS
DOCUMENT NUMBER: PREV200100302761
TITLE: Probing the surface of eukaryotic cells using combinatorial toxin libraries.
AUTHOR(S): **Bray, Mark R.**; Bisland, Stuart; Perampalam, Subodini; Lim, Wai-May; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Princess Margaret Hospital, Ontario Cancer Institute, 610 University Avenue, Rm. 7-117, Toronto, Ontario, M5G 2M9, Canada
gariepy@uhnres.utoronto.ca
SOURCE: Current Biology, (1 May, 2001) Vol. 11, No. 9, pp. 697-701. print.
CODEN: CUBLE2. ISSN: 0960-9822.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 27 Jun 2001
Last Updated on STN: 19 Feb 2002
AB The success of proteomics hinges in part on the development of approaches able to map receptors on the surface of cells. One strategy to probe a cell surface for the presence of internalized markers is to make use of **Shiga-like toxin** 1 (SLT-1), a ribosome-inactivating protein that kills eukaryotic cells. SLT-1 binds to the glycolipid globotriaosylceramide, which acts as a shuttle, allowing the toxin to be imported and routed near ribosomes. We investigated the use of SLT-1 as a structural template to create combinatorial libraries of toxin variants with altered receptor specificity. Since all SLT-1 variants retain their toxic function, this property served as a search engine enabling us to identify mutants from these libraries able to kill target cells expressing internalizable receptors. Random mutations were introduced in two discontinuous loop regions of the SLT-1 receptor binding subunit. Minimal searches from screening 600 bacterial colonies randomly picked from an SLT-1 library identified toxin mutants able to kill cell lines resistant to the wild-type toxin. One such mutant toxin was shown to bind to a new receptor on these cell lines by flow cytometry. Toxin libraries provide a strategy to delineate the spectrum of receptors on eukaryotic cells.

L29 ANSWER 5 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2001:380656 BIOSIS
DOCUMENT NUMBER: PREV200100380656
TITLE: The use of **Shiga-like toxin** 1 in cancer therapy.
AUTHOR(S): **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Department of Medical Biophysics, University of Toronto and the Ontario Cancer Institute, University Health Network,

610 University Ave., Toronto, Ont., M5G 2M9, Canada
gariepy@oci.utoronto.ca
SOURCE: Critical Reviews in Oncology-Hematology, (July-August, 2001) Vol. 39, No. 1-2, pp. 99-106. print.
ISSN: 1040-8428.
DOCUMENT TYPE: Article
General Review; (Literature Review)
LANGUAGE: English
ENTRY DATE: Entered STN: 8 Aug 2001
Last Updated on STN: 19 Feb 2002

L29 ANSWER 6 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2002:93726 BIOSIS
DOCUMENT NUMBER: PREV200200093726
TITLE: Intracellular trafficking of **Shiga-like Toxin** I.
AUTHOR(S): LaPointe, Paul G. [Reprint author]; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Medical Biophysics, University of Toronto, 610 University Avenue, Rm. 7-105, Toronto, ON, M5G 2M9, Canada
SOURCE: Molecular Biology of the Cell, (Nov, 2001) Vol. 12, No. Supplement, pp. 84a. print.
Meeting Info.: 41st Annual Meeting of the American Society for Cell Biology. Washington DC, USA. December 08-12, 2001. American Society for Cell Biology.
CODEN: MBCEEV. ISSN: 1059-1524.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 24 Jan 2002
Last Updated on STN: 25 Feb 2002

L29 ANSWER 7 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2000:211979 BIOSIS
DOCUMENT NUMBER: PREV200000211979
TITLE: **Shiga-like toxin** 1 as a potent purging agent for autologous stem cell grafts of patients with breast cancer, lymphoma and multiple myeloma.
AUTHOR(S): **Gariepy, Jean** [Reprint author]; Bray, M. R.; Patterson, B.; Lim, W.-M.; Perampalam, S.; Keating, A.; Stewart, A. K.; Miller, N.; Banerjee, D.; Belch, A. R.; Pilarski, L. M.; LaCasse, E. C.
CORPORATE SOURCE: Cross Cancer Institute, Edmonton, AB, Canada
SOURCE: Proceedings of the American Association for Cancer Research Annual Meeting, (March, 2000) No. 41, pp. 120-121. print.
Meeting Info.: 91st Annual Meeting of the American Association for Cancer Research. San Francisco, California, USA. April 01-05, 2000.
ISSN: 0197-016X.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 24 May 2000
Last Updated on STN: 5 Jan 2002

L29 ANSWER 8 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1999:511242 CAPLUS
DOCUMENT NUMBER: 131:140494
TITLE: Cytotoxic heteromeric protein combinatorial libraries
INVENTOR(S): **Gariepy, Jean; Bray, Mark Robert**

PATENT ASSIGNEE(S): Ontario Cancer Institute, Can.
SOURCE: PCT Int. Appl., 61 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9940185	A1	19990812	WO 1998-CA1137	19981208
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2222993	AA	19990804	CA 1998-2222993	19980204
CA 2319720	AA	19990812	CA 1998-2319720	19981208
AU 9915530	A1	19990823	AU 1999-15530	19981208
AU 769824	B2	20040205		
EP 1051482	A1	20001115	EP 1998-959689	19981208
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
JP 2002503453	T2	20020205	JP 2000-530599	19981208

PRIORITY APPLN. INFO.: CA 1998-2222993 A 19980204
WO 1998-CA1137 W 19981208

AB Provided is a method for constructing, identifying and using new therapeutic or diagnostic proteins capable of binding to a target cell. The present invention utilizes the concept of using a multi-tasking heteromeric protein toxin, such as **Shiga toxin** or other related ribosome-inactivating protein (RIP), as a mol. template in developing powerful cytotoxic agents having the ability to bind specifically to target cells. By modifying residues affecting only the receptor-binding specificity of the toxin template, it is possible in accordance with the invention to use the toxic A subunit present in all mutant toxins as a mol. search engine in screening combinatorial protein libraries of the toxin's template to find mutant toxins that kill specific cells or cell types.

REFERENCE COUNT: 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L29 ANSWER 9 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 2000:32133 BIOSIS
DOCUMENT NUMBER: PREV200000032133
TITLE: Deletion analysis of the **Shiga-like Toxin** I A chain: Minimum domain for catalytic activity and cytotoxicity.
AUTHOR(S): LaPointe, Paul G. [Reprint author]; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: University of Toronto, 610 University Avenue, Rm 7-105, Toronto, ON, M5G 2M9, Canada
SOURCE: Molecular Biology of the Cell, (Nov., 1999) Vol. 10, No. SUPPL., pp. 107a. print.
Meeting Info.: 39th Annual Meeting of the American Society for Cell Biology. Washington, D.C., USA. December 11-15, 1999. The American Society for Cell Biology.
CODEN: MBCEEV. ISSN: 1059-1524.
DOCUMENT TYPE: Conference; (Meeting)

Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 13 Jan 2000
Last Updated on STN: 31 Dec 2001

L29 ANSWER 10 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1997:542510 CAPLUS
DOCUMENT NUMBER: 127:146836
TITLE: A method for selectively purging CD77+ cells from bone marrows
INVENTOR(S): **Gariepy, Jean**
PATENT ASSIGNEE(S): Ontario Cancer Institute, Can.
SOURCE: PCT Int. Appl., 25 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9729181	A1	19970814	WO 1997-CA77	19970203
W: CA, JP				
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
US 5801145	A	19980901	US 1996-599211	19960209
CA 2245762	AA	19970814	CA 1997-2245762	19970203
EP 879280	A1	19981125	EP 1997-901492	19970203
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
JP 2001500002	T2	20010109	JP 1997-528004	19970203
PRIORITY APPLN. INFO.:			US 1996-599211 A	19960209
			WO 1997-CA77 W	19970203

AB A method for the selective purging ex vivo of CD77 pos. cells from bone marrow prior to autologous transplantation is described. The method involves treating the bone marrow with **shiga toxin** or **shiga-like toxin**-1 to kill CD77+ cells or to remove them by affinity chromatog. The toxin selectively binds to CD77+ cells and not to other bone marrow cells. The method offers a means for curing non-Hodgkin's lymphomas.

L29 ANSWER 11 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 2
ACCESSION NUMBER: 1998:86651 BIOSIS
DOCUMENT NUMBER: PREV199800086651
TITLE: Orientation in lipid bilayers of a synthetic peptide representing the C-terminus of the A1 domain of **Shiga toxin**. A polarized ATR-FTIR study.
AUTHOR(S): Menikh, Abdellah; Saleh, Mazen T.; **Gariepy, Jean**; Boggs, Joan M. [Reprint author]
CORPORATE SOURCE: Res. Inst., Hosp. Sick Children, 555 University Ave., Toronto, ON M5G 1X8, Canada
SOURCE: Biochemistry, (Dec. 16, 1997) Vol. 36, No. 50, pp. 15865-15872. print.
CODEN: BICHAW. ISSN: 0006-2960.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 24 Feb 1998
Last Updated on STN: 24 Feb 1998

AB The interaction of a synthetic peptide representing the C-terminal 27 amino acids of the A1 domain of **Shiga toxin** (residues 220-246) with acidic phospholipid model membranes was characterized by FTIR spectroscopy. This peptide resembles a signal sequence and may

mediate the translocation of the catalytic A1 chain of **Shiga toxin** to the cytoplasm following its retrograde transport to the lumenal compartment of the endoplasmic reticulum (ER). At pH 7 and 5, the peptide underwent a conformational change from random coil to alpha-helix upon addition of negatively charged phospholipids. Examination of the amide II band in the spectrum of the complex at pH 7 and pH 5 showed that in both cases, the N-H groups in the peptide backbone are largely protected from H/D exchange. Using polarized attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FTIR) measurements, the orientation of the alpha-helical portion of the peptide was found to be almost perpendicular with respect to the membrane plane at pH 7. However, at pH 5.0-5.4, the alpha-helix axis was preferentially oriented parallel to the membrane plane. The results suggest that at the neutral pH of the ER lumen, the peptide may insert into the membrane, while at the lower pH levels present in earlier endocytic compartments, the peptide would be less likely to traverse the bilayer. In summary, this putative signal peptide may not be able to cause a significant translocation of the A1 domain of **Shiga toxin** to the cytosol until it reaches the neutral pH of the ER compartment.

L29 ANSWER 12 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 3

ACCESSION NUMBER: 1996:421800 BIOSIS
DOCUMENT NUMBER: PREV199699144156
TITLE: Insertion and orientation of a synthetic peptide representing the C-terminus of the A-1 domain of **Shiga toxin** into phospholipid membranes.
AUTHOR(S): Saleh, Mazen T.; Ferguson, Jim; Boggs, Joan M.; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Dep. Med. Biophys., Univ. Toronto, Ontario Cancer Inst., 610 University Ave., Toronto, ON M5G 2M9, Canada
SOURCE: Biochemistry, (1996) Vol. 35, No. 29, pp. 9325-9334. CODEN: BICHAW. ISSN: 0006-2960.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 10 Sep 1996
Last Updated on STN: 10 Sep 1996

AB **Shiga toxin** is a bacterial protein composed of one A and five B subunits. Its A chain possesses a protease sensitive loop (Cys-242-Cys-261) that is cleaved to produce an enzymatically active A-1 domain and an A-2 fragment associated with its B subunit pentamer. The proposed mode of action of the toxin is linked to its retrograde transport to the ER lumen followed by the translocation of its catalytic A1 chain to the cytoplasmic side of the ER membrane. A signal sequence-like domain (residues 220-246) which constitutes the C-terminus of the A-1 chain precedes a region within the protease sensitive loop (residues 247-258) that contains known and putative cleavage sites. Two peptides corresponding to this C-terminus (residues 220-246) were chemically synthesized to investigate if this signal sequence-like domain can interact with membranes. Such a property may provide a clue to the mechanism of translocation of the A-1 domain across the ER membrane. The first peptide represented the native sequence, which includes a naturally occurring cysteine at position 242 and provided a thiol moiety for the attachment of a spin-label. A second peptide was designed to contain a single tryptophan residue (Ile232Trp) located within the hydrophobic core of the sequence which served as an intrinsic fluorescence probe. The interactions of both peptides with lipid vesicles were analyzed by circular dichroism, fluorescence, and EPR spectroscopy: The peptides lack structure in aqueous buffers and adopted an alpha-helical geometry when bound to negatively charged lipid vesicles. The addition of lipid vesicles to a solution of the tryptophan-containing peptide results in a blue shift in the wavelength of its fluorescence maxima as well as an

increase in fluorescence intensity at 335 nm, suggesting that the hydrophobic core of this A1 peptide relocated to a nonpolar environment. EPR measurements of a proxyl-labeled analog of the peptide (introduced at Cys-242) indicated a decreased mobility of a fraction of the proxyl probe in the presence of lipid vesicles. At pH 7, the membrane-bound probe was completely reduced by ascorbate trapped inside vesicles but only partially reduced by ascorbate added outside the vesicles, suggesting that the C-terminal region of the peptide traversed the membrane bilayer or relocated close to the surface of its inner lipid leaflet. Finally, the peptide was shown to insert into lipid vesicles, causing the release of calcein at a high peptide:lipid ratio. These results suggest that the C-terminal tail of the A-1 chain may anchor this domain into the ER membrane.

L29 ANSWER 13 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 4

ACCESSION NUMBER: 1996:457459 BIOSIS
DOCUMENT NUMBER: PREV199699179815
TITLE: **Shiga-like toxin** purges human lymphoma from bone marrow of severe combined immunodeficient mice.
AUTHOR(S): Lacasse, Eric C.; Saleh, Mazen T.; Patterson, Bruce; Minden, Mark D.; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Dep. Med. Biophysics, Ontario Cancer Inst., Princess Margaret Hosp., 610 University Ave., Toronto, ON M5G 2M9, Canada
SOURCE: Blood, (1996) Vol. 88, No. 5, pp. 1561-1567. CODEN: BLOOAW. ISSN: 0006-4971.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 11 Oct 1996 Last Updated on STN: 11 Oct 1996

AB **Shiga-like toxin**-1 (SLT-1) is a bacterial toxin that kills cells by inhibiting protein synthesis. SLT-1 is composed of one cytotoxic A-subunit and five B-subunits that bind to CD77, a cell-surface glycolipid. In the human hematopoietic system, CD77 expression is restricted to a subset of activated B cells and derived cancers. Here we report that SLT-1 treatment of murine bone marrow ex vivo effectively cures severe combined immunodeficient mice of a human B-cell lymphoma xenograft while sparing normal hematopoietic precursor cells. Flow cytometry results using fluorescein isothiocyanate-labeled SLT-1 B-subunit show the high prevalence of expression of SLT-1 receptors (CD77) in human non-Hodgkin's lymphomas, especially follicular lymphomas. These results suggest the use of SLT-1 for the purging of human bone marrow before autologous bone marrow transplant in the case of CD77+ B-cell lymphomas as just one of many possible uses.

L29 ANSWER 14 OF 16 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 5

ACCESSION NUMBER: 1993:168812 BIOSIS
DOCUMENT NUMBER: PREV199395089862
TITLE: Local conformational change in the B-subunit of **Shiga-like toxin** 1 at endosomal pH.
AUTHOR(S): Saleh, Mazen T.; **Gariepy, Jean** [Reprint author]
CORPORATE SOURCE: Ontario Cancer Inst., 500 Sherbourne St., Toronto, ON M4X 1K9, Canada
SOURCE: Biochemistry, (1993) Vol. 32, No. 3, pp. 918-922. CODEN: BICHAW. ISSN: 0006-2960.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 31 Mar 1993

Last Updated on STN: 31 Mar 1993

AB Shiga and Shiga-like toxins are potent bacterial cytotoxins composed of six proteins: one A-subunit that possesses a toxic N-glycosidase activity and a pentamer of identical B-subunits that anchors the toxin to glycolipids present on mammalian cells. Following their endocytosis through coated pits, a segment of the A-subunit noncovalently associated with the B oligomer is translocated to the cytoplasm where it enzymatically inactivates the protein synthesis machinery. The fluorescence intensity of the single tryptophan residue in the B-subunit is perturbed by pH conditions typically observed in an endosomal compartment, with a sharp reversible transition in fluorescence intensity occurring at pH 4.5. The secondary structure of the pentamer as monitored by circular dichroism is altered by pH conditions lower than 4.5 and greater than 10. However, the conformational change observed under acidic conditions as low as pH 2 does not parallel a loss of receptor binding potential and is reversible, suggesting that the structure of the B-subunit undergoes a second conformational change between pH 4.5 and 2 without a loss of tertiary or quaternary structure. The B-subunit may thus play a role in the translocation of the A chain to the cytoplasm, an event potentially mediated by a conformational change in its structure at pH levels occurring in the endosomal or lysosomal compartments.

L29 ANSWER 15 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:141110 CAPLUS
DOCUMENT NUMBER: 114:141110
TITLE: Serological responses to the B subunit of **Shiga-like toxin** 1 and its peptide fragments indicate that the B subunit is a vaccine candidate to counter the action of the toxin
AUTHOR(S): Boyd, Beth; Richardson, Susan; **Gariepy, Jean**
CORPORATE SOURCE: Dep. Med. Biophys., Univ. Toronto, Toronto, ON, M4X 1K9, Can.
SOURCE: Infection and Immunity (1991), 59(3), 750-7
CODEN: INFIBR; ISSN: 0019-9567
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The B subunit of **Shiga toxin** and **Shiga-like toxin** 1 (SLT-1) and its fragments are potentially immunogenic and may generate protective humoral responses against the action of these toxins. The antibody response was analyzed of rabbits immunized with pure B subunit of SLT-1 or synthetic fragments of the subunit. The immune response to the native B subunit was found largely directed at conformational epitopes. More importantly, rabbits immunized with the B subunit were protected from a lethal challenge with SLT-1, indicating that the B subunit represents an excellent vaccine candidate to counter the effects of **Shiga toxin** and SLT-1 in humans. Polyclonal antibodies against a synthetic peptide corresponding to residues 28-40 of the B subunit neutralized the cytotoxicity of SLT-1 towards Vero cells. This region is thus exposed in the native state of the B subunit. The sequence specificity of other antipeptide antisera also provides clues to the state of folding and assembly of the B subunit. Antisera to synthetic peptides representing the N- and C-terminal regions of the SLT-1 B subunit did not cross-react with native B subunit but strongly recognized denatured forms of the protein. Finally, the monoclonal antibody 13C4 was shown to bind to a discontinuous epitope expressed only on the native form of the protein. These immunol. reagents can be used to probe the conformational state of the B subunit and the holotoxin as it relates to their functional properties.

L29 ANSWER 16 OF 16 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:1843 CAPLUS
DOCUMENT NUMBER: 114:1843

TITLE: Characterization of **Shiga-like toxin** I B subunit purified from overproducing clones of the SLT-I B cistron

AUTHOR(S): Ramotar, Karam; Boyd, Beth; Tyrrell, Gregory; **Gariepy, Jean**; Lingwood, Clifford; Brunton, James

CORPORATE SOURCE: Samuel Lunenfield Res. Inst., Mount Sinai Hosp., Toronto, ON, M5G 1X5, Can.

SOURCE: Biochemical Journal (1990), 272(3), 805-11
CODEN: BIJOAK; ISSN: 0306-3275

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The cistron encoding the B subunit of Escherichia coli **Shiga-like toxin** I (SLT-I) was cloned under control of the tac promoter in the expression vector pKK223-3 and the SLT-I B subunit was expressed constitutively in a wild-type background and inducibly in a lacIq background. The B subunit was located in the periplasmic space, and less than 10% was found in the culture medium after 24 h incubation. Polymyxin B exts. contained as much as 160 μg of B subunit/mL of culture. B subunit was purified to homogeneity by ion-exchange chromatog. followed by chromatofocusing. Crosslinking anal. of purified native B subunit showed that it exists as a pentamer. In gels containing 0.1% SDS the native protein dissociated into monomers. B subunit was found to have the same glycolipid-receptor-specificity as SLT-I holotoxin. Competitive binding studies showed that B subunit and holotoxin had the same affinity for the globotriosylceramide receptor. Evidently, this recombinant plasmid is a convenient source of large amts. of purified SLT-I B subunit, which could be used for biophys. and structural studies or as a natural toxoid.

=> d his

(FILE 'HOME' ENTERED AT 15:52:27 ON 18 NOV 2004)

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 15:53:29 ON 18 NOV 2004

L1 0 SHIGA 5A TOXIN
L2 193 (SHIGA (5A) TOXIN) (S) (RESISTANT OR RESISTANCE OR INSENSITIVE
L3 1 ((MUTATED OR MUTANT) (S) SUBUNIT) AND L2
L4 20901 ((MUTATED OR MUTANT) (S) SUBUNIT)
L5 182 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (5A) TOXIN)
L6 122 (MUTATED OR MUTANT OR VIRULENT) (S) (SHIGA (W) TOXIN)
L7 16 L2 AND L6
L8 10 DUP REM L7 (6 DUPLICATES REMOVED)

FILE 'STNGUIDE' ENTERED AT 16:07:50 ON 18 NOV 2004

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:08:34 ON 18 NOV 2004

L9 2 (MUTATED OR MUTANT) (W) ((VIRULENT OR VIRULENCE OR TOXICITY OR
L10 2 DUP REM L9 (0 DUPLICATES REMOVED)
L11 1 L2 AND L4
L12 125 DUP REM L2 (68 DUPLICATES REMOVED)

FILE 'STNGUIDE' ENTERED AT 16:42:37 ON 18 NOV 2004

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 16:58:30 ON 18 NOV 2004

L13 2725 (MUTATED OR MUTANT) AND ((VIRULENT OR VIRULENCE OR TOXICITY OR
L14 15 L13 AND SHIGA (3N) TOXIN
L15 6 DUP REM L14 (9 DUPLICATES REMOVED)

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     FILE 'STNGUIDE' ENTERED AT 17:03:25 ON 18 NOV 2004
L16             0 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 17:19:55 ON 18
     NOV 2004
L17         32942 RICIN OR ABRIN OR GELONIN OR CROTIN OR POKEWEED (W) ANTIVIRAL (
L18           108 L13 AND L17
L19            41 DUP REM L18 (67 DUPLICATES REMOVED)

     FILE 'STNGUIDE' ENTERED AT 17:31:53 ON 18 NOV 2004

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, WPIDS' ENTERED AT 17:46:35 ON 18
     NOV 2004
L20            15 (SHIGA (W) TOXIN OR SHIGA (W)LIKE(W)TOXIN) AND L13
L21             0 L20 NOT L14
L22             0 L13 AND (GARIEPY J?/AU OR BRAY M?/AU)
L23          1647 (GARIEPY J?/AU OR BRAY M?/AU)
                  E GRIEPY JEAN/AU
                  E GARIEPY JEAN/AU
L24           137 S E3 OR E4
L25           187 S E1 OR E3 OR E4
                  E BRAY M/AU
                  E BRAY MARK/AU
L26            35 S E7 OR E8
L27           217 S L25 OR L26
L28            21 (SHIGA (W) TOXIN OR SHIGA (W)LIKE(W)TOXIN) AND L27
L29            16 DUP REM L28 (5 DUPLICATES REMOVED)

=> l27 and l17
L30             1 L27 AND L17

=> l27 and l13
L31             0 L27 AND L13

=> l30 not l29
L32             0 L30 NOT L29

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